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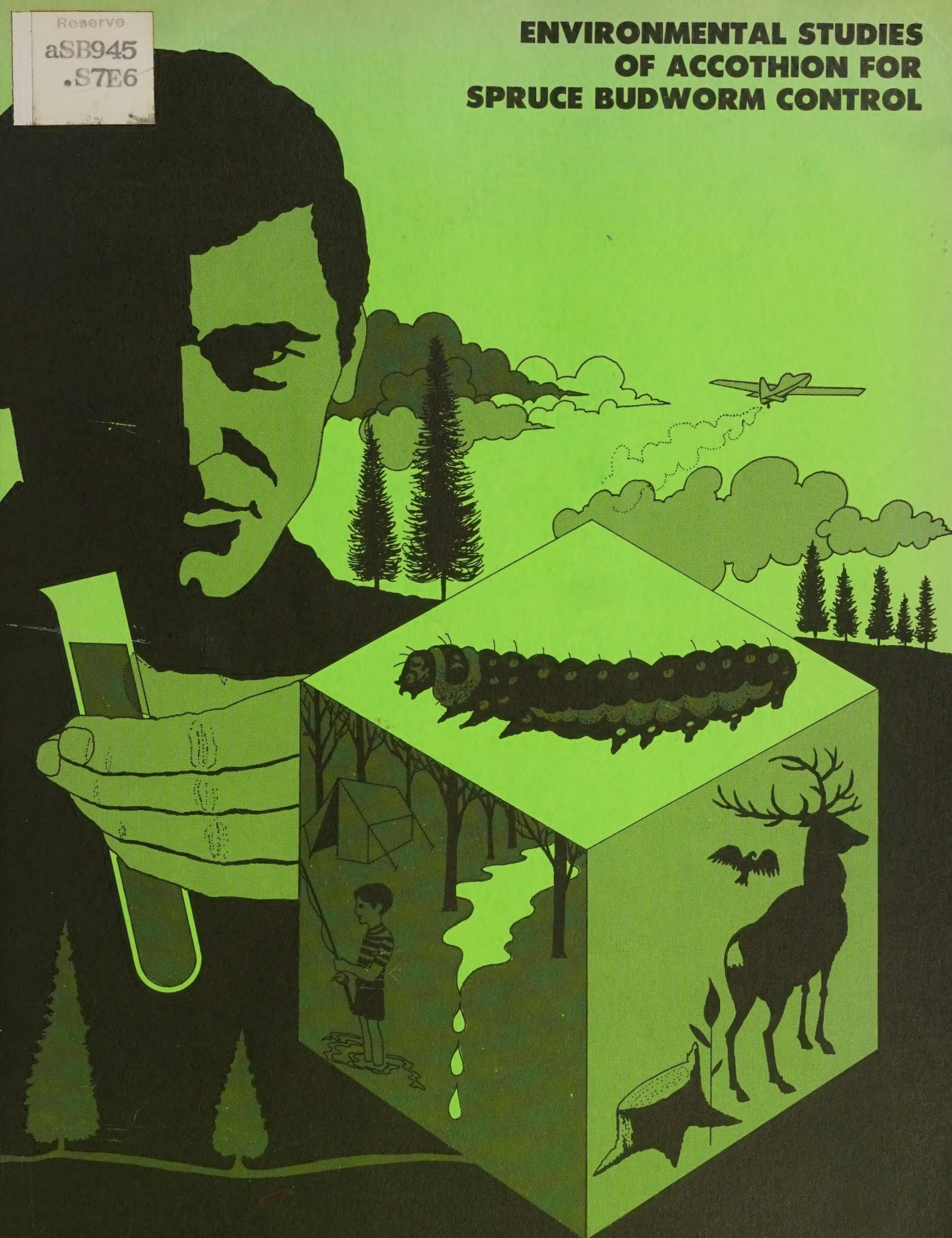
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# ENVIRONMENTAL STUDIES OF ACCOTHION FOR SPRUCE BUDWORM CONTROL



A COOPERATIVE STUDY BY THE STATE OF MAINE,  
U.S. DEPARTMENT OF AGRICULTURE AND U.S. DEPARTMENT OF INTERIOR



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Environmental Studies in the  
Use of Accothion for  
Spruce Budworm Control

by

State of Maine, Department of:

Forestry  
Agriculture  
Inland Fisheries and Game  
Health and Welfare  
Sea and Shore Fisheries

University of Maine

U.S.D.I. Bureau of Sport Fisheries and Wildlife

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February 1971

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This joint report was compiled by a three-man coordinating committee selected by the authors of these special studies. Questions or comments on specific points should be addressed to the author(s). Additional copies of this joint report or general information can be obtained from the coordinating committee chairman.

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ABSTRACT

Application operations were preceeded by extensive planning and pre-cautionary measures. Base was at the Presque Isle Airport with the full operations crew living together as a unit at nearby facilities. The two applications were made in the period May 29 - June 15 inclusive. Direct costs were \$1.194 per acre, addition of indirect costs or overhead brought this figure to \$1.355.

Men assigned to hazardous areas at the airport facility were monitored for exposure to the insecticide chemical. Three blood tests were made on each man for plasma cholinesterase activity, the results showing no toxic exposure of any individual to the insecticide. The total aspect of equipment, drum, and area decontamination was supervised. A recommendation concerning elimination of drum decontamination was made.

Effects of the insecticide treatment throughout the treated area were determined from pre- and post-treatment foliage collections from trees at 86 locations scattered throughout the treatment area. A reduction in survival of 84% can be attributed to the insecticide. Routine surveys of the spruce budworm infestation made annually by Maine Forestry Department personnel were used. The reduction of 84% in survival was not sufficient to alter the course of the infestation in the treated area, for a comparison of egg masses in the 1969 and 1970 surveys showed only a slight reduction in the treated area. Outside the treated area the epidemic budworm infestation expanded in size and intensity. The treatment was successful in preserving much current foliage, especially on those trees most severely damaged in recent years. The treated area was sufficiently improved in overall tree condition that treatment was not recommended for 1971.

Intensive monitoring of budworm populations on five heavily infested plots showed that when the prescribed dosage reached the target tree a substantial amount of current foliage is saved. The prescribed dosage did not effectively suppress populations but left sufficient survivors to produce generally heavy levels of new egg masses. The first application has the greatest impact on foliage protection with relatively little effect on population reduction. Whereas the second application has the greatest impact on population reduction with little effect on foliage protection. The data suggests that a higher dosage in the second application (5th instar) would provide better population control.



The broad spectrum of Accothion is evident from the knockdown of predaceous and parasitic arthropods, including spiders, and parasitic Diptera and Hymenoptera. Spruce budworm parasites killed, include Meteorus trachynotus and Glypta fumiferanae. The spiders and parasitic dipterans were not identified and may also contain species which attack budworm. There were no striking differences in the mortality of budworm larvae harboring parasites at the time of spraying.

Populations of stream invertebrates were monitored by bottom and/or drift sampling in six sprayed and six unsprayed streams. No reductions were detected in the variety of invertebrates collected. Total numbers collected were reduced somewhat, however. The reductions were statistically significant only for the Chironomidae, and these reductions appeared in all sprayed streams. Reductions in other taxa were restricted to one of the six sprayed streams, and it is suggested that this stream may have received an overdose of spray application. The impact of Accothion on aquatic invertebrates appear minor in comparison to some other pesticides, e.g. DDT.

Observations in the spray area were made on reactions to Accothion of a great variety of species of flora and fauna present. Accothion showed observable effects on some insects but not on other insects; nor on plants, birds, minnows in a confined pool, tadpoles, frogs, toads, and salamanders.

Two brooks located inside the spray area and one, serving as a control, located outside the spray area were studied to evaluate possible effects of Accothion upon fish life. No significant short-term changes in pre-spray and post-spray population estimates were evident. No mortality occurred in brook trout held in live cages throughout the study. Fish mortality at the site of blocking seines was high in one of the sprayed brooks and in the control brook. Most of this mortality is believed to have been caused by high flows coupled with spawning movements of many of the minnow species. The second application of Accothion may have contributed to a high mortality of common shiners. Samples of brook trout, white suckers, common shiners, creek chubs, and redbelly dace, will be analyzed for pesticide residues.

Pre- and post-spray bird censuses were conducted in both the spray and control area. Evening grosbeaks were apparently attracted to the budworm infestation area as 196 were recorded in the spray area as compared to 38 in the control area. An intensive carcass search covering an estimated 90 linear miles of tote road within the spray area disclosed no bird mammal or amphibian mortality or signs of aberrant behavior. One dead parula warbler was found in the spray area. A very depressed brain cholinesterase value strongly suggested that death was caused by Accothion. Two applications of Accothion



at a rate of two ounces per acre did not cause significant bird, mammal or amphibian mortality.

Free-flying sapsuckers were used to monitor the effects of Accothion on bird mortality and behavior. Observations of 74 birds were made, before and after spraying, at nine nesting territories in the sprayed area and at four in the unsprayed area. Sapsucker nestlings were banded and placed in nest-blocks, where normal nesting activities continued. Nestlings fledged and lived normally in their home territories. The spray apparently did not cause sapsucker mortality. Some mortality occurred but was probably caused by nest disturbance and predators.

Samples of grass forage, milk, and soil were chemically analyzed and showed the presence of Accothion up to 1.1 ppm. on grass, no presence in milk, and no presence in soil.

## INTRODUCTION

In the fall of 1969 the Maine Forestry Department faced a damaging spruce budworm infestation on over 200,000 acres of spruce-fir forests in south central Aroostook County. The outbreak had been building over the past three years, the situation had been predicted. The budworm problem in the adjacent Canadian provinces was grim also. To the east in neighboring New Brunswick an infestation extended over nearly 3.4 million acres. To the north in Quebec, new infestations were expanding and covered nearly 50,000 acres. A careful evaluation of the biological and economic implications of the Maine outbreak showed that chemical control was the only acceptable alternative.

Recommendation for control action in 1970 was made by entomologists of the Maine Forestry Department. The recommendation was accepted by the Forest Commissioner, Land Managers and the U.S. Forest Service. Control action involved 210,000 acres centered in the Oxbow area of northern Maine (See map in Appendix).

With the phase out in 1967 of DDT for spruce budworm control, we were faced with need for a suitable substitute insecticide. Canadian research and large-scale operational control programs in neighboring New Brunswick, Canada indicated that the insecticide Accothion\* at the rate of 1/8 pound per acre applied twice, was safe in the environment and provided acceptable control. The alternative chemicals were either unsuitable to Maine conditions or not available for use. Voluminous data gathered from studies in New Brunswick by Canadian forestry and other environmental agencies was submitted to the U.S. Department of Agriculture which resulted in granting of a temporary permit for use of Accothion in Maine. The control program was carried out in the period May 29 - June 15.

Knowing that this was the first wide scale use of Accothion in the United States, we made a concentrated effort to monitor it closely. Our objective was to determine the effects on the budworm and other life forms in the forest environment when Accothion was used under operational conditions. All but two of the environmental studies made in connection with the 1970 program are reported here. The field study on Accothion residues in the forest soil is not reported due to

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\* Active Ingredient: O, O-dimethyl O-p-nitro-m-tolyl phosphorothioate



a failure in laboratory analytical procedures to identify the residues. A bioassay using brine shrimp was inconclusive due to insufficient replication in the experiment.

Project funding was arranged on an equal three-way basis. State funds were provided by Legislative action. The legislature also enacted a requested tax by and on all owners in the unorganized townships regardless of where they owned land (the so-called Maine Forestry District Tax). Federal funds were provided through the U.S. Forest Service under the provisions of the Forest Pest Control Act.

## Planning and Application of the Insecticide

Robley Nash, State Entomologist

Maine Forestry Department, Augusta, Me.

### Planning and Public Relations

Extensive planning and preparations took place for months in seeing that all was in complete readiness on the date spraying should start. Readiness is very important so as not to slight any activities and because both the season for budworm control as well as periods with air conditions suitable for spraying are short.

Discussion meetings were held with all agency representatives who were to be involved in the various environmental studies.

A 37-page set of control specifications described all that the spray plane contractor was to supply, his specific procedures, financial and operational responsibilities, and financial penalties in case of infractions.

Public meetings were held to explain full details of the needs, plans, and expectancies of the operation to conservation agencies and groups. These were followed up with similar meetings at Presque Isle just prior to the start of operations to explain procedures at the site. Explanatory articles of the same nature were given the news media.

Pre-spray personal contacts were made with all residents in or within one-quarter to one-half mile of the spray area. This was a repeat procedure of all previous operations. Purposes were (1) to explain all details of the operation and answer any questions and (2) to seek out dairy, poultry, mink, or bee farms, and fish ponds. Also sought out, were all high utility, radio, TV poles or towers. All were marked by erecting beacons and/or on U.S. G.S. maps. Owners of farms and ponds were given follow-up telephone notice just prior to spraying. At pre-spray briefing sessions guide navigators and spray pilots were supplied with these maps and told to shut-off spraying and not to turn over such



farms and ponds, as well as rivers, lakes, and streams. Maine and Federal Aviation Agencies and Loring Air Force Base advised us on avoiding conflicts with aircraft regulations and Air Force operations. Other precautions followed are given under the airport and Human Health sections.

Over-all planning and supervision of the project was by the State Entomologist and his staff. Personnel and assignments for the over-all operation are given in the organization chart on the following page.

The guessing game of setting the date for having materials ready and personnel on hand at Presque Isle at the right time to avoid adding to expenditures was met perfectly. Original estimates could be well made from previous experience with average spring development of the budworm and with past operations. Final date was pin-pointed more closely each day as May advanced from data supplied by field crews daily analyzing the insect's development

#### Airport Facilities and Mixing

As has been usual in large spray operations, the cooperation of other divisions of the Maine Forestry Department and many other agencies was extremely helpful. The Northern Maine Vocational Technical Institute provided meals and a dormitory for housing those involved with the spray application. The State Department of Agriculture certified and sealed the meters involved in loading insecticide into the aircraft. United States Customs and Immigration and Maine State Police cooperated in moving of men and equipment from New Brunswick to Maine. The State of Maine Insurance Department cooperated in bond and insurance matters, Purchasing Office in authorizing hurriedly needed purchases, State Public Health Laboratories in safety matters.

The city of Presque Isle was helpful in various matters; the Maine Public Service Company set marker poles and the Aroostook Airways supplied various airport needs and map-room space.

During the week of May 11th, Maine Forestry Department Fire Control personnel installed cribbing for a large storage tank. On May 20th they set up the mixing and loading equipment and system together with a crew from Forest Protection Ltd. of New Brunswick by whom these items had been prepared and rented. Later they installed meters at three aircraft loading stations for accurate dispensing of the insecticide into spray planes, plus a water supply system from a nearby pond and holding tanks to insure a ready supply of water. Presque Isle city water was not used because it is chlorinated and therefore adverse to the stability of Accothion.

JUNE, 1970, AROOSTOOK COUNTY, MAINE, SPRUCE BUDWORM PROJECT, 210,000 ACRES, AND ACCOMPANYING SURVEYS.  
 BASE-Presque Isle Municipal Airport

OPERATED BY:  
 STATE OF MAINE  
 FOREST SERVICE  
 DIVISION OF ENTOMOLOGY

OPERATING FUNDS:  
 STATE, PRIVATE FEDERAL

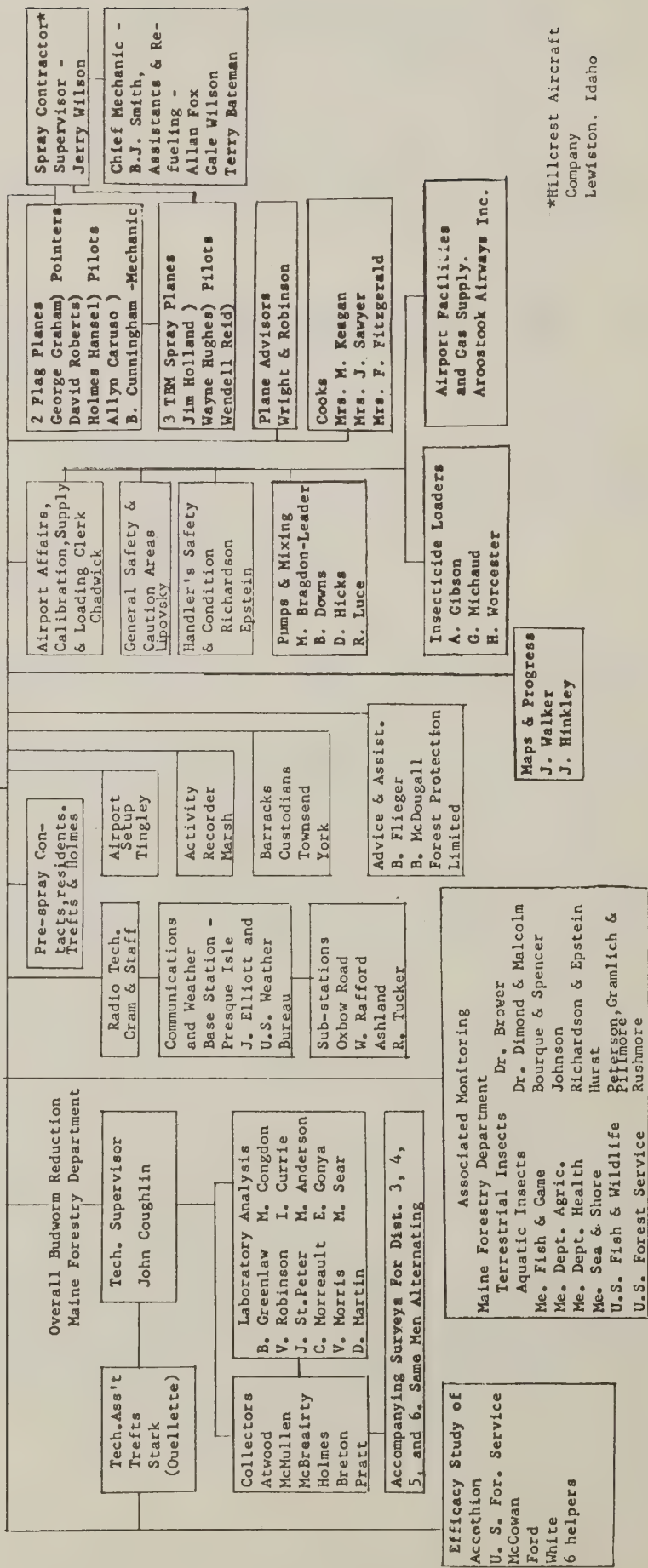
Forest Commissioner  
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 Deputy Commissioner  
 Fred Holt

Business Manager  
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U.S. Forest Service Coordin.  
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- COOPERATION BY:
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  - 2) FOREST PROTECTION LTD.  
 OF NEW BRUNSWICK
  - 3) PRESQUE ISLE CITY MANAGER  
 FIRE-POLICE DEPARTMENT
  - 4) NORTHERN MAINE VOCATIONAL  
 TECHNICAL INSTITUTE

Project Director  
 R. W. Nash, State Entomologist



\*Hillcrest Aircraft  
 Company  
 Lewiston, Idaho



All others assigned to the airport arrived May 27th. Together they completed final preparations on the afternoon of May 29, including installation by Department radio technicians of the all-important radio communication system between airport headquarters, spray planes, flag (or guide) planes, and ground crews. The three TBM spray planes and pilots were obtained through a 37-page contract with Hillcrest Aircraft Company of Lewiston, Idaho. Two Cessna 172 aircraft and pilots for guiding the spray planes were contracted from Bar Harbor Airways, Ellsworth, Maine.

Spray planes were dispatched May 28th to Dunphy Airstrip, New Brunswick, for calibration by Forest Protection Ltd. at their headquarters since they had all previous operational experience in recent years with the insecticide.

On May 28th and 29th, the insecticide arrived in 48.8 gallon drum lots. It was immediately placed in a roped-off storage area marked, "No Trespassing" and provided with the 24-hour police protection in effect for all airport facilities throughout the operation.

Mixing and loading crews were thoroughly briefed beforehand in handling the insecticide as described in the section on Human Health. Gasoline was brought to the spray planes by truck for refueling when reloading with insecticide. Fire extinguishers were provided around all pumps and planes and a manned city fire truck was present during actual operations.

The insecticide Accothion emulsifiable concentrate was known as 8EC because each gallon contained 8 pounds of actual Accothion. Mixing involved 10.4 gallons 8EC with enough water to make 100 gallons of water emulsion. Each batch of water emulsion was mixed fresh just before loading into planes for application. A batch of 1875 gallons of emulsion required for each 3-plane load involved transferring 195 gallons of 8EC (4 drums of 48.8 gallons each) to a measuring tank on top of the mixing tank. A separate pump was provided for this and the measuring tank had an outside gauge. At the same time 1680 gallons of water was pumped and metered into the mixing tank. Just prior to loading the planes, a valve at the base of the measuring tank was opened to allow the concentrate to flow down into the mixing tank to the water. A line from the pump used to load the aircraft led back to the mixing tank to provide agitation and mixing of the 8EC and water for a few minutes, then the line was opened to pump the mixture



State of Maine forestry officials brief USDA and USDI representatives on the proposed 1970 suppression project (fall, 1969)



State and federal officials planning a strategy to monitor the effects of Accothion on the forest environment.



directly into the spray planes. The lines to the planes were fitted with 2-inch Buckeye "Dry-break" couplings (two to each plane) to eliminate drippings when planes were loaded and couplings disconnected.

### Application Rates

Two applications of Accothion were planned, timed to coincide with certain instar development in the life of the spruce budworm caterpillars. Requirements under Temporary Permit No. 241-Exp-50G issued by the Federal Government for the project called for the first application to begin when larval development had reached the peak of the third instar, the second application after development had reached the peak of the fourth instar.

As May advanced, development of the budworm caterpillars was continually followed by the Maine Forestry Department's Portage laboratory (Details are given in Coughlin's report on this laboratory's work.) The peak of the third instar was reached on May 29th. Spray application began that evening. The first application was completed on June 7th and one load was applied to start the second application. The peak of the fourth instar had passed by that date. The second application was completed on June 15th. (See Coughlin's report, Figure 1).

The application rate of 2 ounces of actual Accothion in 0.15 gallons of emulsion per acre was achieved by the three spray planes each having 625 gallons per load, or a total of 1875 gallons. This team load was applied to a spray block of 12, 500 acres.

### Releasing Spray Blocks

Decisions to spray and method of controlling the spray planes were through the mobile flag-plane system devised by Forest Protection Ltd. For the one team of spray planes, two flag (Cessna) planes were used each carrying an experienced navigator, one of which was assigned responsibility as chief.

Weather reports radioed to the air base from department of weather or fire lookout stations in the area started before daybreak and continued every half hour until spraying stopped. Reports started again in the evening as soon as



AUTHORIZED

Project direction and communications were centralized at the Presque Isle Airport. Police protection was provided by the City of Presque Isle.



Insecticide mixing plant and TBM aircraft at Presque Isle Airport.



winds showed any sign of being sufficiently low in velocity. Reports gave wind direction and velocity, temperature, dew point, and relative humidity, fog conditions, haze and possibility of rain. Navigators flying over the area checked low altitude conditions and with the weather reports radioed to them made their calculations. The chief then gave the decision to spray or not to. Likewise, the navigators made the decision when conditions demanded spraying stop. At times, the spray pilots with their experience were of help in radioing their observations. The U. S. Weather Bureau at Caribou and Loring Air Force Base gave good help in our planning by giving advance forecasts as well as precautions on when to expect shower activity.

Spraying was done more often from daybreak to 7 a.m., less often in the evening because winds did not subside in time to operate. However, evening spraying is acceptable because once winds drop and low air turbulence subsides to the point where spraying can start, conditions improve to the better.

In the mobile system of controlling spray planes, basic guidance was from the navigators in the flag-planes. Navigators established each line and course to be flown according to prepared maps and the terrain. Flight altitude was roughly 700 feet for the flag planes. Spray planes flew at 100-150 feet as a team of three with the lead plane lined up on, and following the two flag planes. The second and third spray planes were 440 feet to one side and behind the lead and second planes respectively. Radio communications allowed navigators to tell spray pilots of needed course corrections, when to turn spray on or off, and when to turn to spray return lines of the block being treated. Spray blocks were approximately 3 miles by  $6\frac{1}{2}$  miles (12,500 acres) and were usually laid out with, and always flown on, the long axis north-south. Spraying proceeded back and forth until the block was completed with twelve passes or swaths by the three planes to complete a block.

## Effects of Accothion on Human Health

Ernest M. Richardson, State Chemist, Pesticides  
Maine Department of Health and Welfare, Augusta, Me.

### Objectives

The State Public Health Laboratory assisted the Maine Forestry Department in monitoring the health aspect of exposure of personnel to Accothion. State Chemist, Ernest Richardson was assigned as Health Officer and as the informal assistant to the Safety Officer. In addition to human monitoring of exposure to insecticide, it was his responsibility to supervise any necessary decontamination and to act as liaison with Gould Memorial Hospital in preparation for any poisoning emergency.

### Cholinesterase Monitoring Study

Twelve men who were to work constantly in the restricted area were monitored for cholinesterase enzyme inhibition, which is the physiological indicator of exposure to an organo-phosphate compound such as Accothion. These men included mixers, loaders, pump operators, area supervisor, radio dispatcher, one plane mechanic, safety officer, and health officer. A plasma cholinesterase base line was established for each man prior to arrival of the insecticide concentrate. A second test was run after one full week of spraying and a third and final test conducted after completion of the entire project.

Blood was drawn at Gould Memorial Hospital at a cost to the project of \$1.00 per sample. Analysis for cholinesterase activity was done by the Health Officer in a field laboratory set up near the airport. The data collectively showed no detectable change in enzyme activity due to exposure. No deviation was seen that could be attributed to anything other than normal daily variation in cholinesterase activity. The data of the study is shown in Table No. 1.



Table 1

Cholinesterase Activity of Project Personnel Working with the  
Organo-Phosphate Insecticide Accothion

Name	Project Job	Plasma Cholinesterase Activity		
		Base line	pH. Units First check	Final test
Manley Bragdan	Pump Operator	1.021	1.341	1.410
Richard Luce	Pump Operator	1.172	1.210	1.372
Bruce Downs	Mixer	1.055	1.377	1.568
Donald Hicks	Mixer	1.188	1.370	1.353
Albert Gibson	Loader	1.320	1.210	1.300
Harold Woorston	Loader	0.952	1.113	1.142
Gillie Michaud	Loader	0.568	0.705	0.685
Alan Fox	Plane Mechanic	0.833	0.945	*
James Elliott	Radio Dispatcher	1.093	1.145	1.368
John Chadwick	Airport Supervisor	0.755	0.900	*
Louis Lipovski	Safety Officer	1.017	1.018	*
Ernest Richardson	Health Officer	0.831	0.951	1.035

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\* Absent from final test

### Personal Protection

On the site at the airport, the personal health of the men working with the insecticide was overseen by the health officer and/or the project safety officer.

Clothing: All personnel working in the restricted area were required to wear rubber protective clothing, to include rubber rain suit, gloves, boots, and a full-face respirator. Items were purchased for the men locally. Reasons for full rubber dress were twofold; first, to prevent dermal absorption of the insecticide and secondly, to avoid ruining personal clothing in the event that a spill required decontamination, the chemical mixture for which is quite caustic and would destroy clothing and shoes.

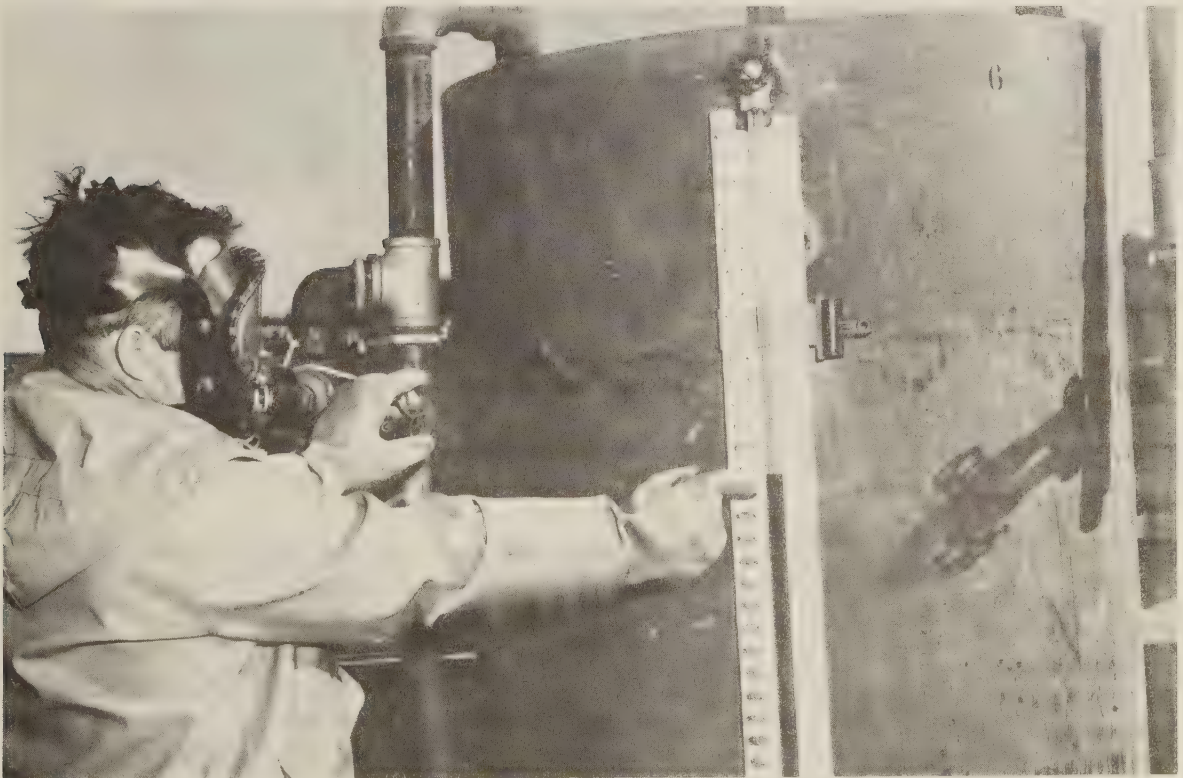
Emergency Medical Backup Support: As part of the overall safety precautions in the handling of organo-phosphate poisons, arrangements were made to have the necessary antidotes on hand at Gould Memorial Hospital in Presque Isle. These chemical antidotes included atropine sulfate (a common hospital item), and 2-pralidoxime iodide (2-PAM or Protopam), purchased by the hospital from Ayerst Laboratories, New York, N.Y. The hospital staff was briefed verbally on the diagnosis and treatment of organo-phosphate poisoning and the staff and emergency team were shown a medically oriented film on the same subject. The orientation resulted in a medical team prepared to handle phosphate poisoning emergencies.

Modification to Canadian Apparatus: It was found that the Canadian mixing tank gave off a considerable amount of strong fumes of Accothion during the mixing process. These fumes were coming from the 24 inch top viewing port and from a 4-inch vent at one end of the tank. It was felt that exposure to these fumes was dangerous and put the mixer in a needlessly hazardous condition regardless of his protective clothing and respirator. The top viewing hole was thus sealed with a lid and rubber gasket (truck inner tube) and a 4-inch by 10-foot extension of iron pipe was made into the 4-inch vent at the end of the tank. These precautions completely eliminated the fume hazard to the mixer and surrounding ground personnel.

### Decontamination Procedures

Personal: It was necessary to provide facilities for personal decontamination in the event of a spill and for





Personnel working in the insecticide restricted area were required to wear rubber rain suits, gloves, boots, and a full face respirator.

decontamination of gloves and boots after each man finished work in the loading pit or concentrate storage area. Two 30 gallon trash cans were purchased for this purpose, one being placed directly adjacent to the concentrate storage area and one centrally located on the loading flight line. Each was equipped with a long handle brush and filled with decontamination solution consisting of two pounds of washing soda, two quarts of chlorox bleach, and one half cup of liquid detergent concentrate (Joy).

Spills: Accidental spillage either on the runway during loading or in the concentrate storage area was decontaminated immediately using the washing soda-bleach-detergent mixture from the decontamination stations mentioned above. Ten-quart pailfuls of the decontaminant were poured onto the spill and the solution scrubbed into the contaminated area with the long handled brushes. Only three minor spills occurred, which offered no problem.

Drum Decontamination: Each 55 gallon Accothion drum contained about one pint of concentrate after the pumping and draining process was complete. Decontamination was performed after draining by the addition of one pound of washing soda, 2 quarts of Chlorox bleach,  $\frac{1}{4}$  cup of Joy detergent and 10 gallons of water. The bung was then replaced and tightened and the drum turned on its side. The drum was rolled and agitated frequently over a 12 hour period to break down chemically any remaining Accothion on the inner surfaces of the drum. After twelve hours or more the bung was removed and the drum drained into a dug sump hole. The drum was then set aside for a final rinse. When about forty drums were awaiting a final rinse, each drum was inverted and set on another and thoroughly rinsed with copious amounts of water under 200 pounds pressure from booster lines of the duty fire truck. After draining, the container was turned right side up, the bung replaced and its exterior also thoroughly washed with this high pressure water stream. At this point the Accothion concentrate drum was considered adequately clean and decontaminated.

Lye is the strongest decontaminant for organo-phosphates. However, its use is hazardous due to its extreme caustic properties. In that Accothion falls in the "slightly toxic" category according to comparative dermal and oral LD-50 values for rats in a publication by Arthur Gall, Maine's Extension Specialist in Pesticide Safety, it was decided that a washing soda-bleach-detergent mixture would be safer to handle and dispose of, and yet do an adequate job of decontamination.



### Conclusions and Recommendation

Data from the human monitoring program showed that with the precautions of special clothing, respirators, and decontamination taken as part of the overall safety program, there was no exposure of personnel to the organo-phosphate severe enough to show any decrease in plasma cholinesterase activity over and above normal daily variation. Healthwise, with regards to exposure to Accothion, each member of the team left the project in as good health as when the project was initiated.

It is felt by this pesticide analyst that if the same type of project is initiated again, a different arrangement should be made for disposal of drums. Decontamination of these drums was easy and considered complete. However, as a result of the decontamination process there was large amounts of foul looking material being poured into an earthen sump to be absorbed into the earth in due time. Had this dumping of the decontaminant been viewed by the onlooking public, there could have been some serious concern and adverse publicity. It is hereby recommended that in the future, drums not be decontaminated at the site, but rather as part of a purchase contract of the insecticide, that the company agree to take back the empty drums. In this way the airport facility and any groundwater or surface drainage nearby would not be contaminated with foreign chemicals in the form of either the toxic or detoxified insecticide or decontaminating agents.

Effects of Accothion on the Spruce Budworm  
(Over-all Project Effectiveness)

John Coughlin, Entomologist  
Maine Forestry Department, Augusta, Me.

Objectives

Entomologists of the Maine Forestry Department undertook several obligations in connection with the 1970 spruce budworm project: 1. To follow the seasonal development of the insect in order to time the various surveys and the spray treatment itself. 2. To determine the effectiveness of Accothion in controlling the insect population and protecting the foliage. 3. To follow the course of the infestation throughout the threatened area. 4. To provide experienced laboratory staff and facilities for studies by U. S. Forest Service personnel (described elsewhere). Since the 1970 work was only a part of a program of annual assessment and surveys of the spruce budworm by the Maine Forestry Department, a considerable reservoir of experience and facilities was available. Similar work goes on every year and is expected to continue as long as the problem continues.

Methods

To follow the development of the insect, a location well within the treatment area was chosen for accessibility and high insect population. On 13 May and every day or two thereafter foliage was collected from the mid-crown of fir trees at this point and brought to the Portage Laboratory. Technicians at the laboratory removed the budworm from the foliage, classified their development, and plotted the results. Other survey collections coming into the laboratory were compared with those plotted to be sure that the point chosen to follow development was representative of the general area to be treated.



As the time for treatment approached, another location, just outside the treatment area, was also followed for development. After treatment started, development in the treated area was retarded and erratic and most information then came from just outside the spray area. Development of the spruce budworm in the Oxbow-Masardis area is outlined in Figure 1.

To determine the effectiveness of the insecticide in controlling the insect, a series of before and after foliage collections were made. Prior to the start of the field season, 125 locations within the treated area and 193 locations outside were listed. Within the treatment area these locations were along existing trails and woods roads well scattered throughout the entire area. Field crews were sent to as many locations as time permitted prior to treatment.

At each point, five 15-inch long branches were taken from the mid-crown of each of five co-dominant fir\* trees using sectional aluminum pole pruners with cloth bags attached just below the cutting head. All 25 branches, bundled in groups of 5, by tree, were placed in a plastic bag and taken to the laboratory. The collection site was assigned a number, plotted on maps, and the 5 sample trees marked with plastic flagging to facilitate re-sampling. At the laboratory, locally hired and trained workers examined each sample branch by branch and recorded the larvae found.

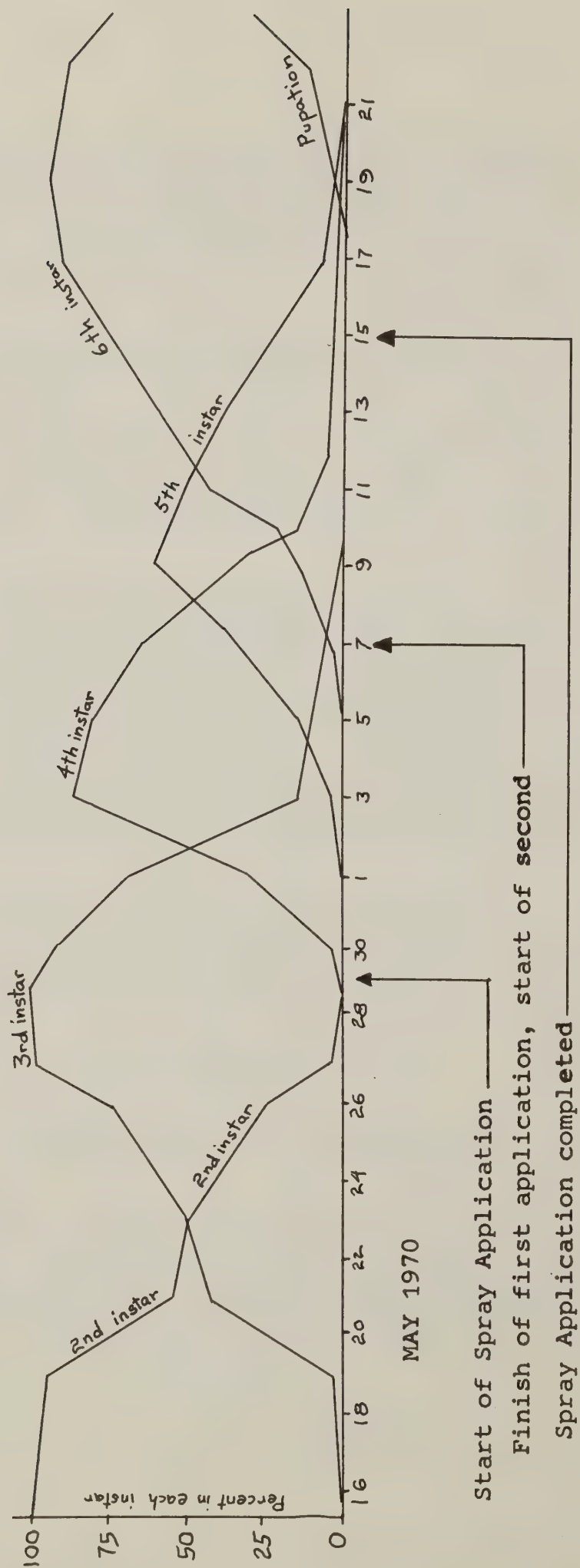
When the insecticide treatment started, field crews were shifted outside the treatment area and made identical collections, working from prepared lists of locations and working out from the periphery of the treatment area. Within the treatment area, a number of 6x6 cloth drop sheets were placed to facilitate observations of spray effect. Two locations were selected and 3 sheets were placed at each location, approximately 2 chains apart along a line perpendicular to spray plane flight lines. These drop sheets were similar to and supplemented similar projects by other agencies.

Following the completion of the spray operation, as many as possible of the pre-spray sample locations were revisited. Normally, the same crew visited each location and sampled the marked trees as before. At the laboratory the samples were examined and results compared with the pre-spray samples.

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\* Red Spruce was sampled instead of fir at four locations. The data were not considered separately but added to the fir samples.

FIGURE 1. DEVELOPMENT OF SPRUCE BUDWORM IN 1970 (DATA FROM OXBOW AND MASARDIS)





Beginning in August, a third survey was made, visiting all listed locations within and outside the treated area and many unlisted locations as well. As before, five 15-inch long branches from each of five co-dominant trees were brought from each location to the laboratory. This third survey sampled the egg masses laid by the 1970 spruce budworm adults and indicated the locations where larval feeding will occur in 1971. These data were plotted on maps and compared with 1969 data to indicate the course of the infestation from one generation to the next.

As soon as budworm feeding was completed, aerial observations were made to record defoliation. Later, in August during the egg mass survey, field crews noted and recorded defoliation at each sample point. Current (1970) defoliation, previous defoliation, and estimates of tree recovery were all recorded and later plotted on maps. A "hazard rating" for each location was then calculated considering the above three factors and the chance for further damage indicated by the egg mass numbers.

### Results

A total of 86 before and after "paired" collections were made within the treated area. (Figure 2) Outside the treated area a similar series of 25 before and after paired collections were made.

Grouping all 86 collections within the treatment area, a total of 34,260 larvae were found in the pre-spray samples. The same trees sampled after spraying yielded 3125 surviving larvae and pupae. Similarly, outside the treatment area 3186 larvae were found in the first series of collections; 1840 were found later on the same trees. Comparing the survival in treated areas with that outside, a reduction in survival of 84.2% can be attributed to the Accothion application (Table 1).

Other effects of the insecticide were noted on the morning of treatment and two days later by observing budworm and other insects on the drop sheets and in the general vicinity. On the morning the area was treated, numerous budworm larvae could be seen hanging by silken threads and dropping to earth. Examination of foliage revealed both sluggish disoriented larvae and others apparently still healthy. On the drop sheets were many dead and dying larvae as well as some apparently healthy specimens. Two days after the second spray application, a re-examination of the drop sheets indicated many dead and dying budworm

FIGURE 2.

LOCATION OF PRE- AND POST-SPRAY SAMPLES  
IN THE TREATED AREA (Maine Forestry Dept. Data)

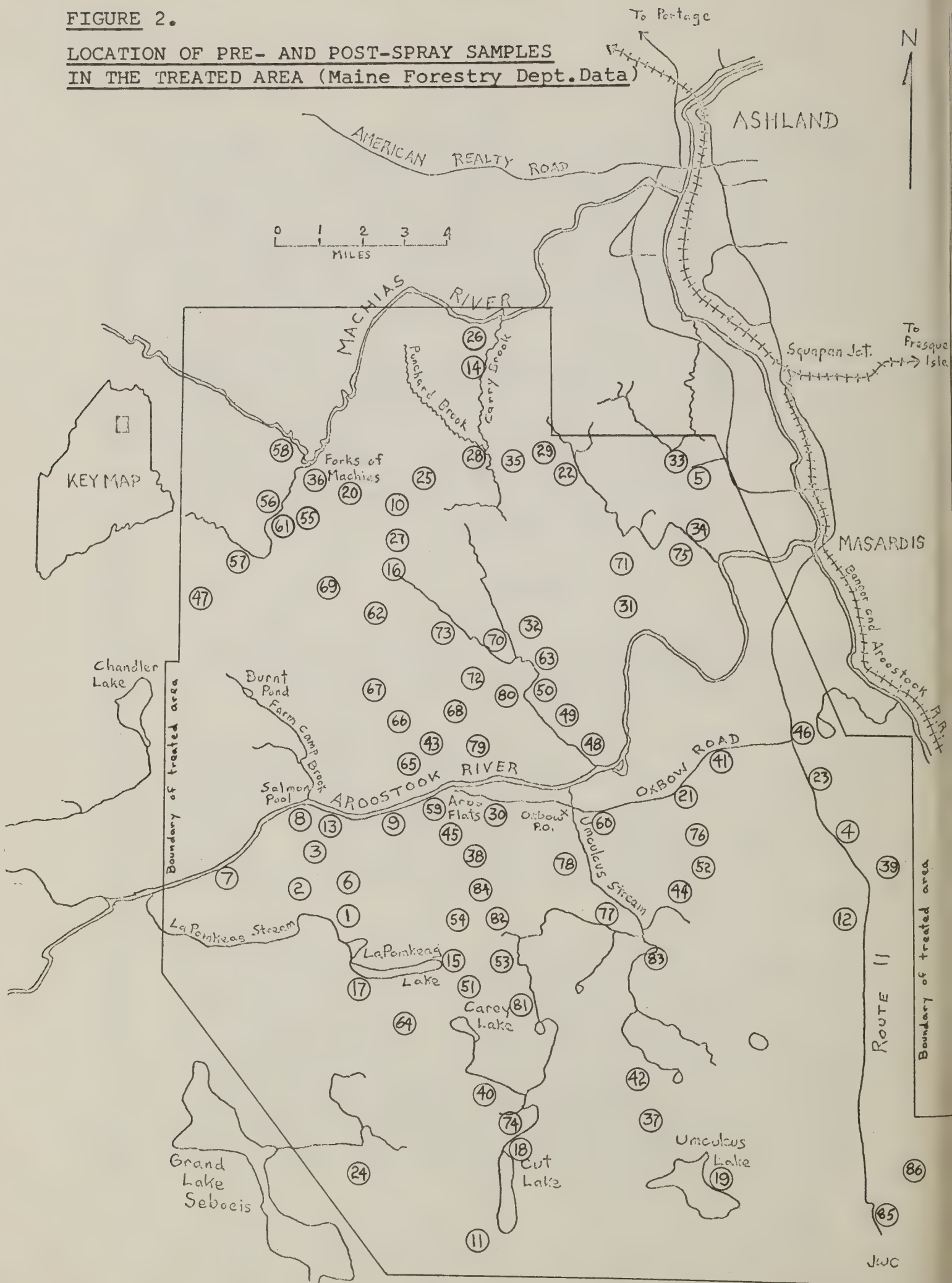




TABLE I

SUMMARY OF 1970 ACCOTHION TREATMENT  
AGAINST SPRUCE BUDWORM

TREATED AREA  
(86 LOCATIONS)

UNTREATED AREA  
(25 LOCATIONS)

Total Budworm  
before treat-  
ment

34,260

3,186

Total Surviv-  
ing budworm  
after treat-  
ment

3,125

1,840

Survival

.091

.577

Percent reduc-  
tion in survival  
by spraying

$$= \frac{.577 - .091}{.577} \times 100 = 84.2\%$$

as well as other insects. Many live budworm collected from the drop sheets subsequently died. While the drop sheets gave no quantitative data, they provided good sites for general qualitative observations.

observations of defoliation from the air and from the ground are combined in Figure 3. A feature of the 1970 defoliation was the total lack of any discernable pattern. The area involved is far from uniform with hardwood ridges, cutting operations going on presently and in the past, and old burns. In general, while there was heavy current defoliation over extensive areas, the areas that were in the worst condition in 1969 seemed to be the most improved in 1970. It was generally believed that the more open foliage of severely injured trees allowed greater penetration of the insecticide. It was also observed that generally the tops of the trees experienced the greatest improvement. Throughout the area it was observed that the forest was in generally improved condition and that there was much new foliage in 1970. This is in general agreement with Canadian experience where protection of current year's foliage is a major goal.

Surveys during the larval period, the aerial survey for defoliation, and the egg mass survey all showed an enlargement of the infestation during the 1970 season. Not only is the defoliated area larger than shown in Figure 3, but a new epicenter seems to have arisen in the Cross Lake-Madawaska Lake area northwest of Caribou. (Figure 4).

Collections of fir foliage from 105 locations within the treatment area and 218 locations outside the area were examined during the egg mass survey. Egg mass numbers within the treated area decreased slightly from an average of 1.440 per 15-inch fir sample tip in 1969 to an average of 1.245 in 1970. Outside the treated area, egg mass numbers more than doubled from an average of .171 per 15-inch fir sample tip in 1969 to an average of .400 in 1970.

The proximity of these infestations to the Canadian border should be noted since extensive and expanding infestations were noted in both Quebec and New Brunswick and Maine is geographically and biologically part of the continuing budworm problem throughout this area.

### Discussion

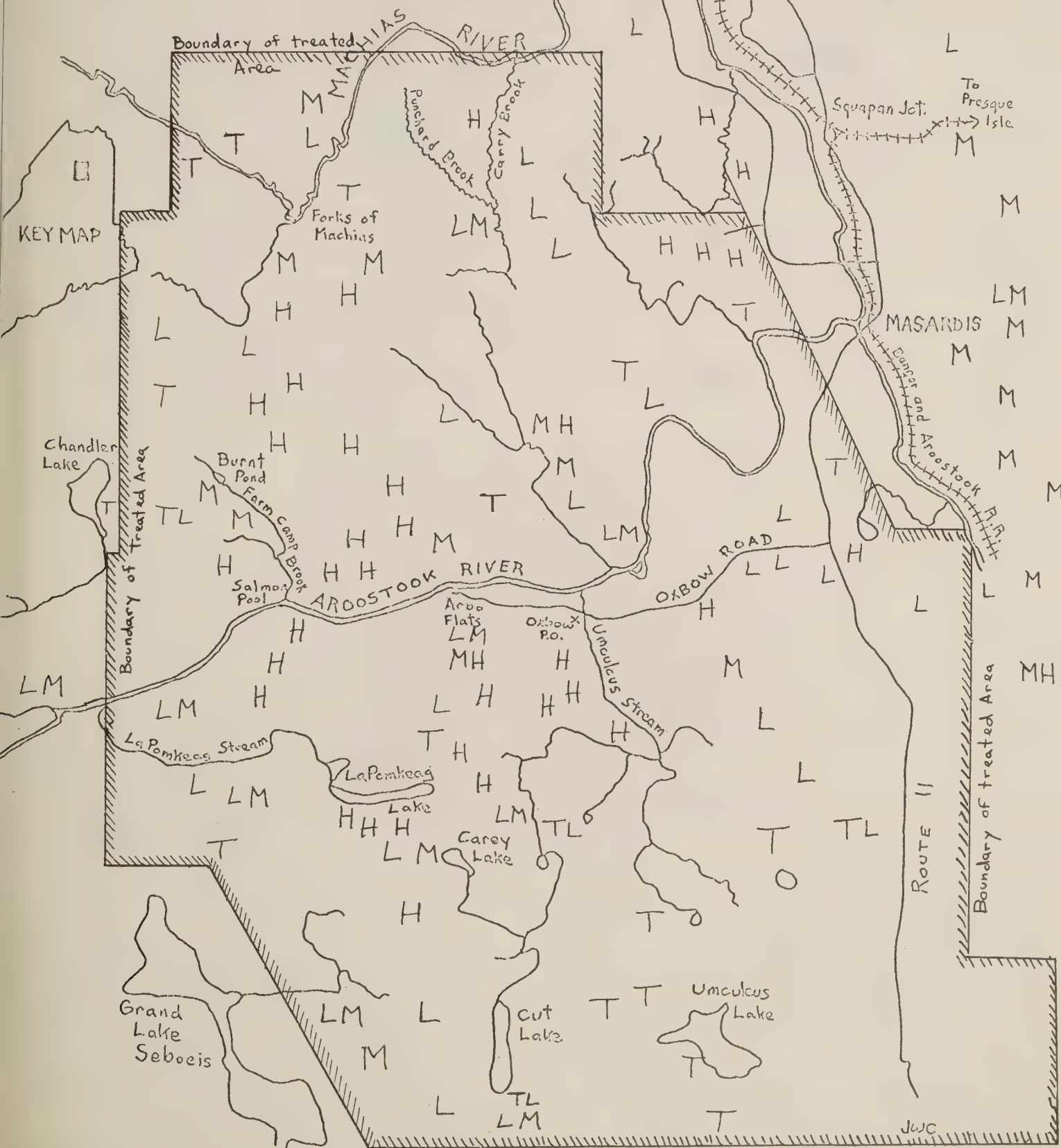
It would appear that the 1970 treatment with Accothion was successful in keeping the infestation from becoming worse in the treated area. Control of the order of 84%, while



FIGURE 3. DEFOLIATION IN THE TREATED AREA, 1970

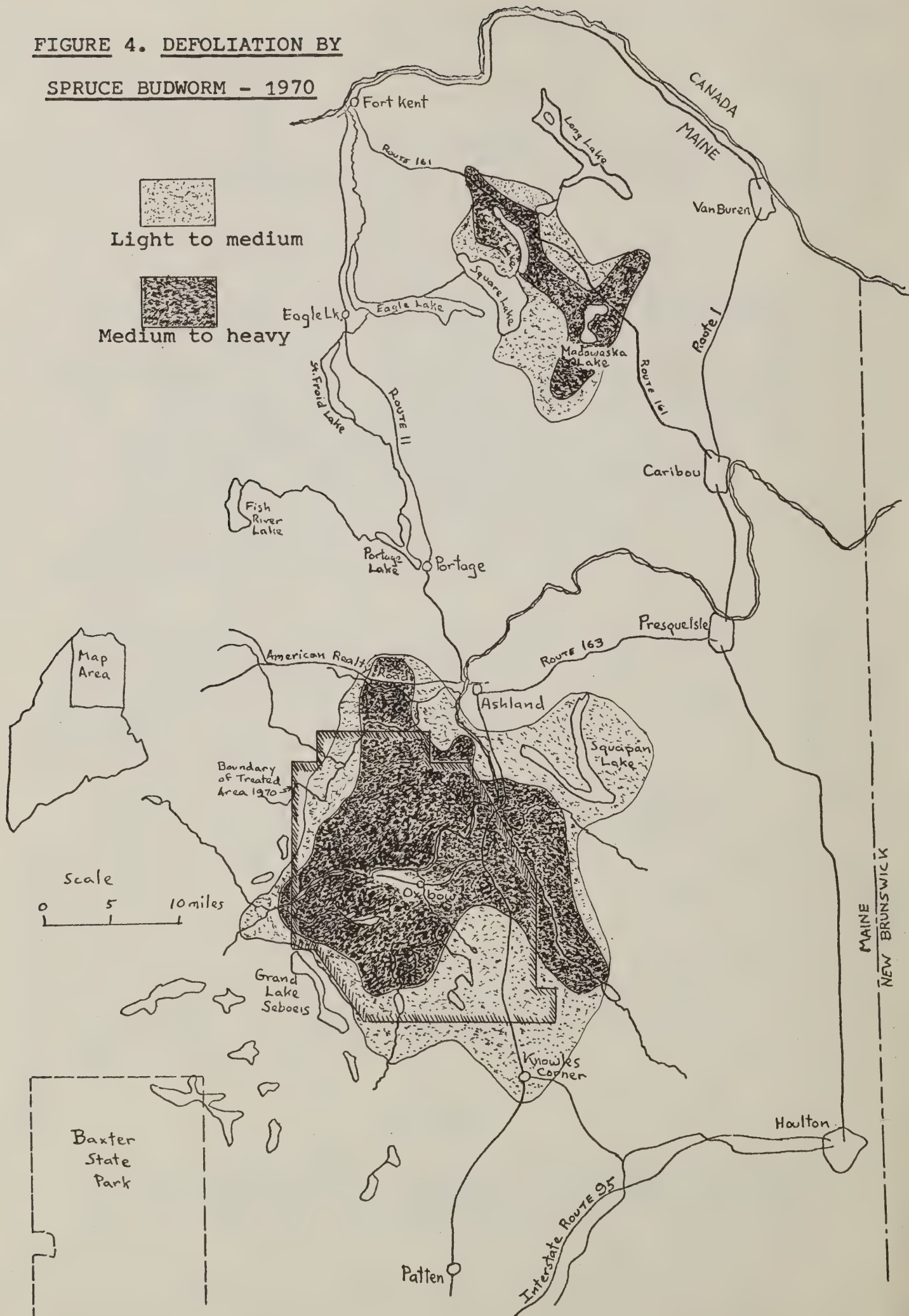
T	Trace	1-5% Def
TL	Trace to light	
L	Light	5-25% Def
LM	Light to medium	
M	Medium	25-65% Def
MH	Medium to heavy	
H	Heavy	65-95% Def

0 1 2 3 4  
MILES



**FIGURE 4. DEFOLIATION BY**

**SPRUCE BUDWORM - 1970**





far short of the degree needed to reduce the spruce budworm population, did seem to be sufficient to prevent increase. While it is unfortunate that experience and methodology do not permit accurate measurements of foliage protection, there was agreement among the experienced personnel present that a high degree of foliage protection had indeed been achieved - so much so that later recommendations were that no budworm control be undertaken for 1971.

Another year's damage from further budworm feeding appears to be tolerable, especially since the 1970 insecticide application gave some relief to the most severely affected trees. The 1971 damage is expected to be severe and action would seem to be needed the following year to prevent widespread mortality.

The problem is clearly becoming worse, for the Oxbow infestation enlarged considerably and the new epicenter arose northwest of Caribou. With present technology it would appear necessary that emphasis be shifted from population reduction to foliage protection as needed, with the overall goal of keeping the trees alive. This would be directly in accord with experience in nearby New Brunswick where the same methods and insecticide have had several years of use.

### Conclusions and Recommendations

It appears to be entirely possible to keep the forest alive with the material and methods used in 1970. The lack of any high degree of population reduction indicates that more frequent treatment will be necessary to achieve this goal than was formerly necessary with DDT in Maine. Since the problem and its treatment are now so closely in accord between Maine and New Brunswick, it is desirable that even closer relationships exist in the future. A better or more effective material than Accothion would be desirable but until or unless one becomes available it is clearly desirable to refine the use of this material hoping for not only foliage protection but population reduction as well.

Effects of Accothion on the Spruce Budworm  
(Intensive Investigations on Small Plots)

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and

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Objectives

The objective of this work was to precisely measure the effectiveness of this insecticide in controlling the budworm. Three measurements of efficacy were taken: 1) The response of budworm populations to known amounts of spray deposit reaching infested trees or clumps of trees; 2) the amount of foliage protected from defoliation by known quantities of spray deposit; and 3) the infestation recovery rate as indicated by the egg population at the start of the first generation following spraying.

The techniques we chose to obtain these measurements closely resembled those used by the Maine Forestry Department (see Coughlin's report). A major difference was in the system we chose to select sampling points. Also our scheme for monitoring population control was extremely intensive by comparison. By using similar techniques we hoped to provide a better understanding of the over-all projects results as reported by Coughlin.

The planning and execution of this work were carried out by the U.S. Forest Service in cooperation with the University of Maine. The Maine Forestry Department provided field laboratory facilities and the personnel necessary to handle and examine the massive samples of foliage.



### Methods

The intensive studies of spray deposition, budworm population reductions, foliage protection, and infestation recovery rate were carried out on five rectangular plots in the more heavily infested portions of the Accothion spray area (see map in Appendix). Maine Forestry Department data on expected budworm population levels were used to locate the five study plots where the insect would be particularly abundant.\* One long side of each rectangular plot was located on a road, providing access, and all but plot IV were oriented so that the long axis of the plot was perpendicular to the flight lines of the spray planes. No more than one plot was located in any single 12,500 acre spray block.

Two additional study plots, outside the spray area, were chosen to serve as unsprayed controls. While these were substantially infested with the budworm, population levels were much lower than in the treated plots. Since the population levels between sprayed and unsprayed plots were not comparable, limited use was made of the data from the latter.

A sampling scheme of completely randomizing sample trees was considered but the forest type and other circumstances made such a scheme completely impractical. Instead, a system of sampling trees along a straight line was chosen.

Preparations of the plots involved marking and clearing a trail and selecting and marking study trees. The trail started at one corner of the rectangular plot at a point of access, proceeded in a straight line to the middle of the opposite long side, then returned diagonally across the plot to the other corner on the proximal long side. This produced  $1\frac{1}{2}$  miles of straight trail but with an angle at its mid point. Fifty balsam fir trees were selected as study trees along the trail and numbered 1-50; twenty spruce were similarly selected and numbered 51-70. Red spruce was selected most often, but where the species was unavailable black spruce or white spruce was used. The high mortality of fir in the interior of plot IV made it necessary to locate more than half of the study trees along a dirt access road that formed one long side of the plot.

Because of the patchy distribution of fir and spruce within the plots, study trees were not at all evenly spaced along the lines. Rather, most occurred in clumps of which two to several were selected and marked. The analysis of data was

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\* Coughlin, J. 1969. The Spruce Budworm in Maine, 1969. Division of Entomology, Maine Forestry Department. Processed, 17 pages.

made on the basis of means of clumps of trees where the individual trees were sufficiently close together that the spray input was essentially uniform. Study trees separated from each other by longer distances were considered as a "clump" containing a single tree.

Criteria for selecting study trees were an open crown and a size, 30-60 feet, allowing removal of mid-crown branch samples with a pole pruner. Since spray deposit cards were to be placed in the open on the ground beside each tree, it was usually necessary to remove vegetation on one side providing a direct opening to the sky.

The degree of opening available for card placement was rated for each tree as follows:

A - opening equivalent to removing several codominant trees; usually trees given an A rating were adjacent to roads or clearings.

B - opening equivalent to removing 1 or 2 codominant trees.

C - a direct opening to the sky existing, but considerably smaller than that produced by removing one codominant crown.

D - no direct opening to the sky.

The great majority of study trees were adjacent to A and B - rated openings, and no differences related to degree of opening were noted in the amount of spray reaching cards placed in these two types of situation. Spray readings on cards in C and D - rated openings were decidedly below average, however, since much was intercepted by the interfering canopy above. Readings of spray deposit for tree clumps were made, therefore, by averaging the deposits only of the cards placed in the higher rated openings of that clump, as these best represented the spray dosage applied to that local area.

Cards used for spray assessment were 3 by 5 inches white, Chromecoat cardboard. Each was stapled at the corners to a slightly larger plywood sheet for exposure to the spray. Cards were placed on the plots immediately before treatment to avoid wetting from rain or dew, and were retrieved within an hour or two after treatment. Spray droplets were counted on 50 sq. cm., about two-thirds of the card, using a microscope, and the size distribution of samples of the droplets measured with an ocular micrometer. Conversion from number and size of droplets to amount of spray received per acre was done after calculating the size on cards of a series of droplets of known volume prepared in the Entomology



Department at the University of Maine. This was confirmed by colorimetric readings of the quantity of dyed spray eluted from aluminum plates exposed alongside deposit cards in the field. A more detailed description of the procedures used to calculate the actual amount of spray can be given by the junior author on request. The procedure was adopted from those used by Chemical Control Research Institute, Ottawa, Canada.

Measurements of spruce budworm populations (larvae and eggs) and defoliation on the study of trees were made with the sampling procedure long used by the Maine Forestry Department. This involved counting the numbers of the insects on five, 15-inch branch tips pruned from the mid-crowns of each study balsam fir. Two branch tips was the sample size established for spruce in an effort to reduce the large task of searching the pruned samples for the insects. Each study tree was sampled 4-5 days before the first application and about 10 days following the second application, the prespray and post-spray budworm population estimates respectively.

Two 5-tree clumps, selected at either end of each study plot, were sampled in early August to estimate the rate of egg mass deposition of the new budworm generation. This provided an evaluation of the infestation recovery rate.

Defoliation of the study trees was estimated in August at the time of the egg mass survey. Feeding of the target generation of the budworm was completed at that time. Estimates were made of the percentage of current needles removed from the shoots by budworm feeding. This was accomplished by viewing in detail several individual shoots against the sky from the ground and with a cursory general view of the remainder of the tree. With practice, an observer becomes adept at defoliation estimations, giving highly reproducible results, although they may differ slightly from those made by other observers. These remarks apply only to fir. Defoliation is much more difficult to estimate on spruce. The needles on the latter are closely appressed to the shoot presenting a poor profile against the sky. Because of lack of confidence in the estimates, spruce defoliation data are not used in this report.

The degree of defoliation of a tree is often not uniform within a crown. Unless there is an extremely heavy budworm population, removing all the current foliage of a tree, there is usually markedly heavier feeding in the upper half of the crown. Because of this and because spray effects may have increased the difference in defoliation between crown levels, separate records were made for both the upper and lower crown halves.

A one-quart sample of the mixed spray was taken at the airport from each load applied to the spray blocks containing the study plots. These samples were sent immediately to the Biochemistry Laboratories of the University of Maine for gas chromatographic analysis of the contents. This step was taken to provide assurance that the study plots were treated with the recommended concentration of Accothion.

## Results

Spruce budworm populations were reasonably uniform on the five study plots located within the spray area. Total larvae from 70 trees per plot, fir and spruce, sampled prior to spray application ran from 2800 to 3200. The average number of prespray larvae per 15" twig on the five intensively sampled plots was 10.1 on the fir and 10.4 on the spruce. The average from 10 nearby sampling points used in the extensive survey by the Maine Forestry Department was 18.4 on fir. The rather large difference is a reflection of the two different schemes to select sample trees. These plots were probably representative of the more heavily infested portions of the total area sprayed.

Spray deposit, however, varied considerably within and between plots, leading to differing budworm population levels and defoliation levels after treatment. The full data for the study plots are voluminous and are not included in this report. Complete data are on file in the Entomology Department, University of Maine and may be seen on request. Summaries and relationships between variables from selected data will be used in this section to illustrate the conclusions drawn from the work.

A brief summary of data arranged by plot means or totals is presented in Table 1. The deposit of spray of 3 oz. listed for plot I is an estimate, not the direct measurement, made for the following reason. The chemical analysis of the spray mixture applied to that block in the first application determined it to contain 4.9 percent Accothion, about one half the recommended concentration (Appendix). While the counts of number of droplets on deposit cards suggested that the plot received a full two ounces of spray in the first application, this was reduced to one ounce, because of the half-strength spray applied. This added to the two ounce dosage recorded in the second treatment produced an estimated average total dosage of 3 oz./acre on that plot. Because of the use of a diluted spray concentration on the first application on plot I, adjusted estimates of spray deposit were used for all further analyses.



Table 1. Summary of Accothion efficacy data (fir only) arranged by plots.

	Prespray Popula- tion	Mean <sup>1/</sup> spray deposit	Mean <sup>2/</sup> defolia- tion		Postspray popula- tion	Survi- vors per twig	Percent <sup>3/</sup> control
Plot I	2364	3 oz.	38	29	519	2.1	78
II	2723	4 oz.	36	28	267	1.1	90
III	2922	1+ oz.	47	47	409	1.6	86
IV	2567	4 oz.	31	41	190	.8	93
V	2155	2 oz.	71	78	1101	4.4	49
Control							
I	1250		50	--	1050	4.2	24
II	650		66	--	850	3.4	0

1/ Ounces of actual Accothion per acre; sum of two applications

2/ Estimate for upper crown (left) and lower (right) crown respectively.

3/ No adjustment for natural control

The spray deposit on plot III is listed as 1+ because of a recorded average of 1 oz./acre deposited in the first application, but with no record of the deposit in the second application, because of failure to place the deposit cards in the field in time before the treatment. Total deposit data are therefore not available, limiting use of the remaining data from that plot. We may presume, however, that unless the second treatment greatly exceeded the recommended 2 oz./acre, the plot received less than the recommended total dosage because the first application was low.

Plot V received poor coverage in both applications, totalling only half the recommended amount of Accothion. This plot was located, however, within one half mile of the spray area boundary where below average coverage can be expected. This study plot was established before it became necessary to drop the 12,500 acre spray block bounding on the north.

Plots II and IV received the recommended total dosage of Accothion, but in different ways. Plot IV received very uniform coverage of about 3 oz. in treatment 1 and 1 oz. in treatment 2. In plot II, coverage in both treatments was erratic. While it averaged 2 oz. in both applications, some tree clumps were missed in one or both treatments, and others overdosed. The great variability in dosages received by different clumps in plot II made these data very





Pole pruners with an attached basket were used to collect 15-inch twig samples from the upper mid-crown of fir and spruce trees.



Women examined the twig samples for spruce budworm. Eight women were stationed at the Portage Lake field laboratory and four were stationed at Cross Lake.



useful in one of the analyses of results described later.

Where the recommended dosage of Accothion was applied, plots II and IV, a decrease in budworm numbers of about 90 percent was achieved. This estimate of reduction has not been corrected for the amount of natural mortality occurring simultaneously, an estimate of which is usually taken from unsprayed control plots. In the present experiment, the control plots provided erratic results of natural budworm declines, one showing an increase in numbers. These are believed to provide a poor representation of events in the more heavily infested experimental plots. The extensive studies of Morris (1963)<sup>2/</sup> suggest that we should expect a 40-50% natural decline in epidemic budworm populations within the period encompassed by our pre- and post-spray collections. Natural population reductions based on the extensively sampled plots averaged 42.3 percent. (See Coughlin's report).

Defoliation on plots II and IV averaged 31 and 36 percent in the upper crown. There are no data available to provide a comparison of the observed level of defoliation with what would be produced by similar levels of budworm populations in the absence of spraying. Previous experience suggests that the level would be 80-90 percent.

Statistical analysis of the data in Table 1 involved paired t tests of comparisons of means. Several of the variables, spray deposit and budworm population counts were transformed to the log 10 (x+1) form to attempt to stabilize mean-variance relationships. These transformations were maintained in all subsequent analyses. All statistical tests were made only at the 95% probability level.

The first spray application was significantly heavier in plot IV and significantly lower in plot II than in the other plots, the other three plots indistinguishable. In the second application, a significant difference was demonstrated only in plot II, which received the heaviest treatment. With the combined, total application as listed in Table 1, only plot IV was significantly different. Plot II data, while the average approached that of plot IV, were much more variable due to uneven application, explaining why a significant difference was not noted.

In the defoliation column, plot V showed the only significant difference between plots in defoliation in the upper crown. With lower crown defoliation, plots I and II were not significantly different from each other, nor were plots III and IV. All other differences reached levels of significance.

Among the survival data, plot V was different from the remaining plots and plot I was significantly higher than plot IV. In

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<sup>2/</sup> Morris, R. F. 1963. Mem. 31, Ent. Soc. Canada, p. 19

percent control, only plot V could be demonstrated as significantly different.

Table 1 provides data only for the fir sample trees. Control of the budworm on spruce was considerably lower in each plot. The percent control figures for spruce were 71, 57, 71, 65, and 23 respectively for plots I-V.

Egg mass survey data for the five plots are presented in Table 2, together with a repeating of some material from Table 1 for comparative purposes. There is relatively little similarity between the degree of control on a specific plot in 1970 and the forecast for populations in 1971, except in the case of plot V, where control was poorest and egg mass counts highest by far. Since moth dispersal occurs during oviposition one would not necessarily expect a good fit on small plots. The infestation recovery rate was high; Table 1 shows that the small plots have a high budworm population again. Coughlin's extensive survey data also showed a high rate of infestation recovery. His ratio of post spray egg mass densities to pre spray densities was .865. However, in unsprayed areas the ratio was 2.355.

One may comment on the overall success of the 1970 Accothion project using the data presented in Tables 1 and 2, assuming that the five plots are representative of the total spray area. The goal of foliage protection was apparently reached satisfactorily. Defoliation on those plots adequately treated was reduced to 30-40 percent from an expected 80-90 percent. Defoliation of the former level can probably be tolerated by fir trees for many years without tree mortality.

The other goal, budworm population reduction, was not satisfactorily realized. With budworm population reductions reaching no higher than 93 percent at best, there were sufficient survivors to produce generally heavy levels of egg masses. Such a situation resembles agricultural pest control where, in spite of heavy kill of insects every year and achieving a goal of product protection, the insect populations remain perpetually in outbreak. This result, up to the present, has generally been considered unacceptable in forest insect control in the United States.

#### Functional Relationships Between the Measured Variables

The greatest benefit, perhaps, from the data collected in the present study are derived from examining functional relationships between the variables studied, e.g. between spray dosage received by a tree clump and the resulting mortality or the resulting degree of defoliation. This allows prediction of effects to be expected if dosages greater or lesser than 4 oz. of Accothion/acre were to be applied. Evidence of this type



is not provided by the simple summation of data, as in Table 1.

Simple regression analyses, using some variables transformed as discussed earlier, were run between all possible pairs of variables as listed in Table 1 and separately for each plot. Least square regression lines were calculated, with 95% confidence limits, and with t tests used to determine significant relationships and for differences between slopes when the different plots were compared.

Table 2. Egg mass collections on balsam fir from experimental plots following Accothion application, with comparative data on budworm control

	Egg masses/25 twigs <u>1</u> /	Expected popula- tions 1971	Survi- vors/twig 1970	Postspray larval popu- lation
Plot I	33	Medium-Heavy	2.1	519
II	39	Heavy	1.1	267
III	26	Medium-Heavy	1.6	409
IV	45	Heavy	.8	190
V	108	Heavy	4.4	1101
Control I	49	Heavy	4.2	1050
II	9	Light-Medium	3.4	850

1/ Mean of two samples

The results of these analyses can be summarized briefly. The dosage of Accothion in the first application was significantly related to degree of control and to defoliation on at least some of the plots, particularly plots I, II and V. The relationship between deposit in the first application and number of budworm survivors is presented in Figure 1, and between that application and defoliation, upper crown, in Figure 2. This spray application appeared, therefore, to influence both objectives of the spray, foliage preservation and budworm population reduction; evidence is presented later suggesting the greatest effect on foliage preservation.

The Accothion deposits in the second application showed significant regression relationships only with survival data, as shown in Figure 3, and not with defoliation estimates. This suggests that the importance of the second application was in

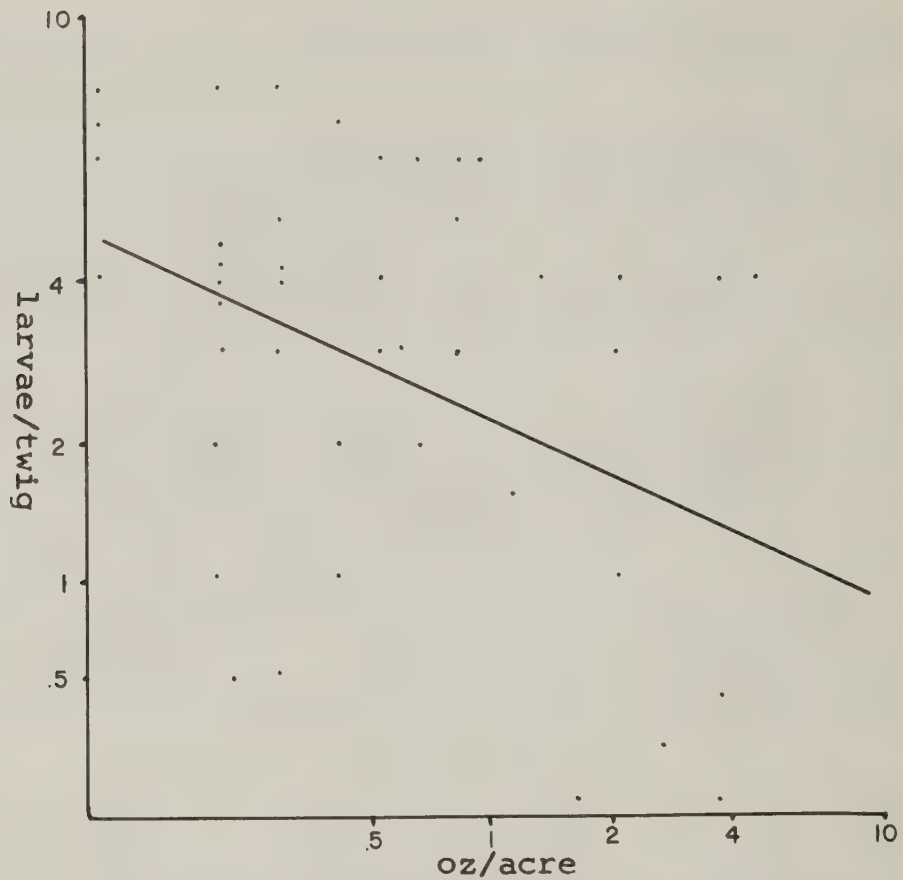


FIGURE 1. Relationship of dosage in first application (X) to numbers of spruce budworm survivors (Y).  $r = -.44$  Plots I and V.

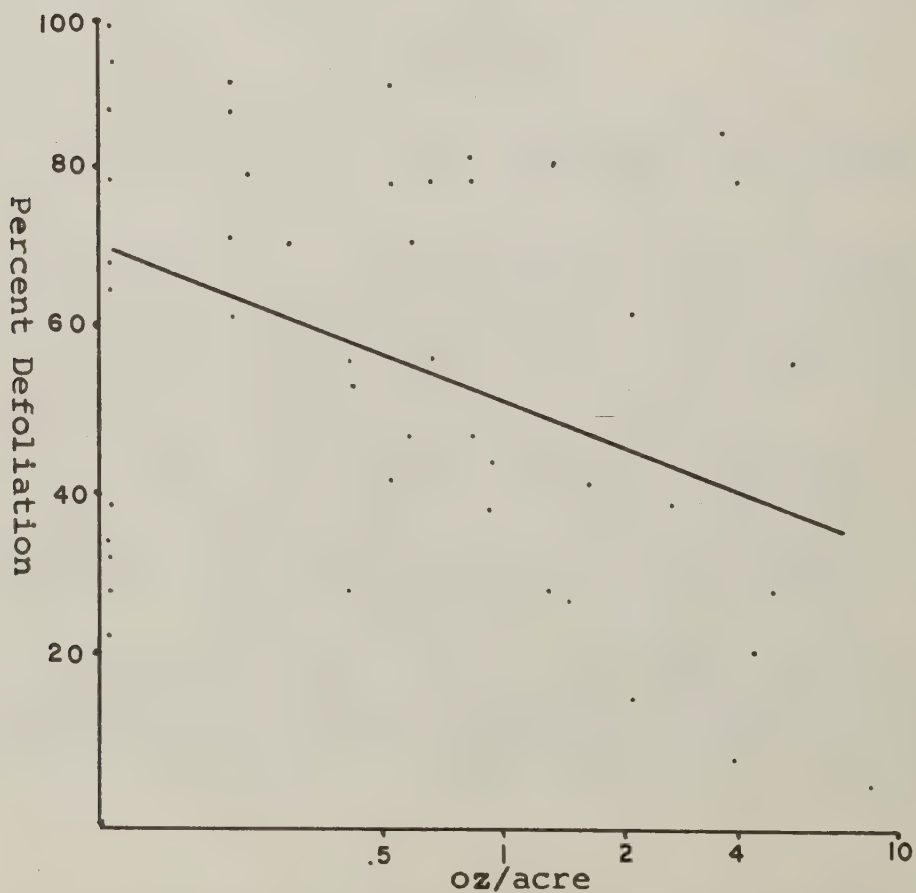


FIGURE 2. Relationship of dosage in first application (X) to percent defoliation (Y).  $r = -.45$  Plots II and V.



population reduction and not foliage preservation. This follows the reported Canadian experience with similar applications of Accothion.

The sum of two applications of spray showed significant relationships with all the other variables on most plots. The relationship with survival is shown in Figure 4 and with defoliation, upper crown, in Figure 5.

Relationships of spray deposit to number of survivors are used in the figures since these relationships were slightly stronger than to percent control. Numbers of survivors and percent control were, of course, strongly related since both were computed from the same population counts. Correlation coefficients between the two variables were above .80. Therefore, conclusions drawn from the relations of spray deposit to survivors apply also to percent control, but in inverse fashion.

In the same sense, only relationship between spray deposit and upper crown defoliation are presented, however, the relationships to lower crown defoliation were similar and actually were statistically stronger. Defoliation in the upper crown is believed to have more importance in terms of tree mortality and damage.

One additional relationship can also be mentioned, that between number of survivors and degree of defoliation, either upper or lower crown. Correlation coefficients varied between .54 and .71 (mean .62) for the various plots. This shows reasonably good agreement between two, independently measured indexes to the effectiveness of the applications of Accothion. Confidence in the results presented is promoted.

A study of the variation in some of the relationships discussed above brought out a point of interest. In the plotted relationship between total spray deposit and number of survivors in plot II, two trends rather than a single one appeared evident. This is illustrated in Figure 6. On investigating further, it was found that all the points but one in the right hand group, the circled points in Figure 6, represented tree clumps that received good spray coverage in both spray applications. All but two points in the left hand group, the uncircled points, on the other hand, received virtually no coverage in the first application, and all of the total dosage was received in the second application. The plotting of these data then, as two trends rather than a single trend, allowed additional conclusions to be drawn on the relative contributions of the first and second treatments alone to budworm population reduction.

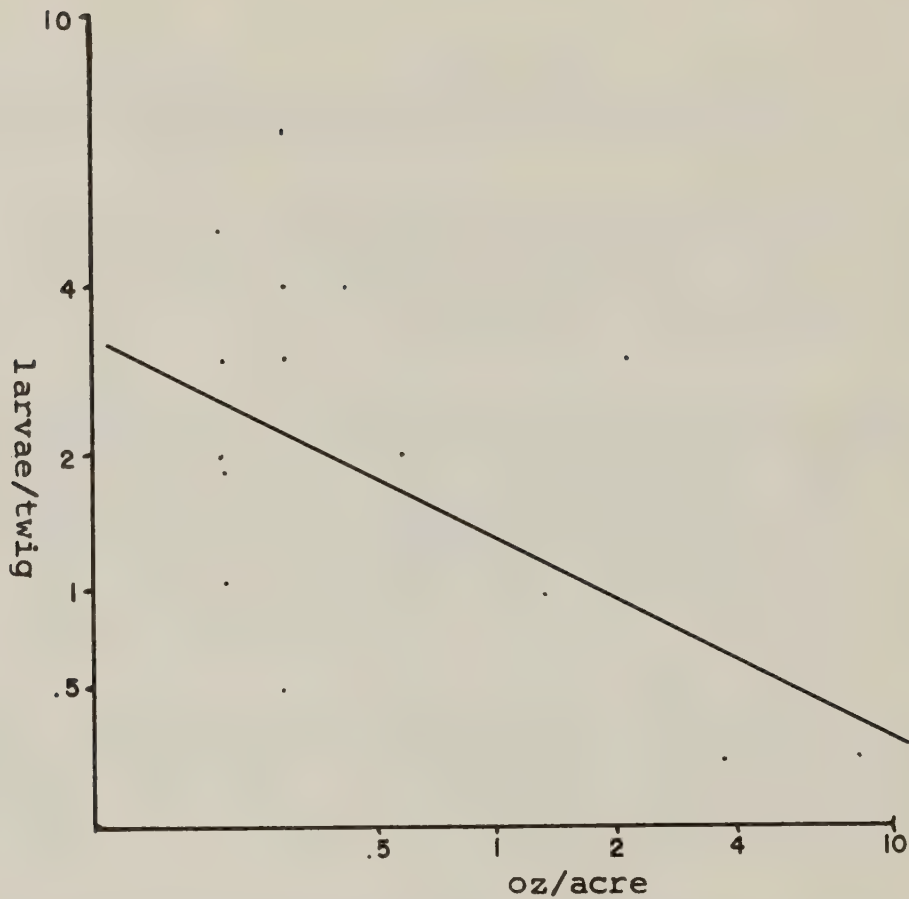


FIGURE 3. Relationship of second application (X) to numbers of spruce budworm survivors (Y).  $r = -.51$  Plot I.

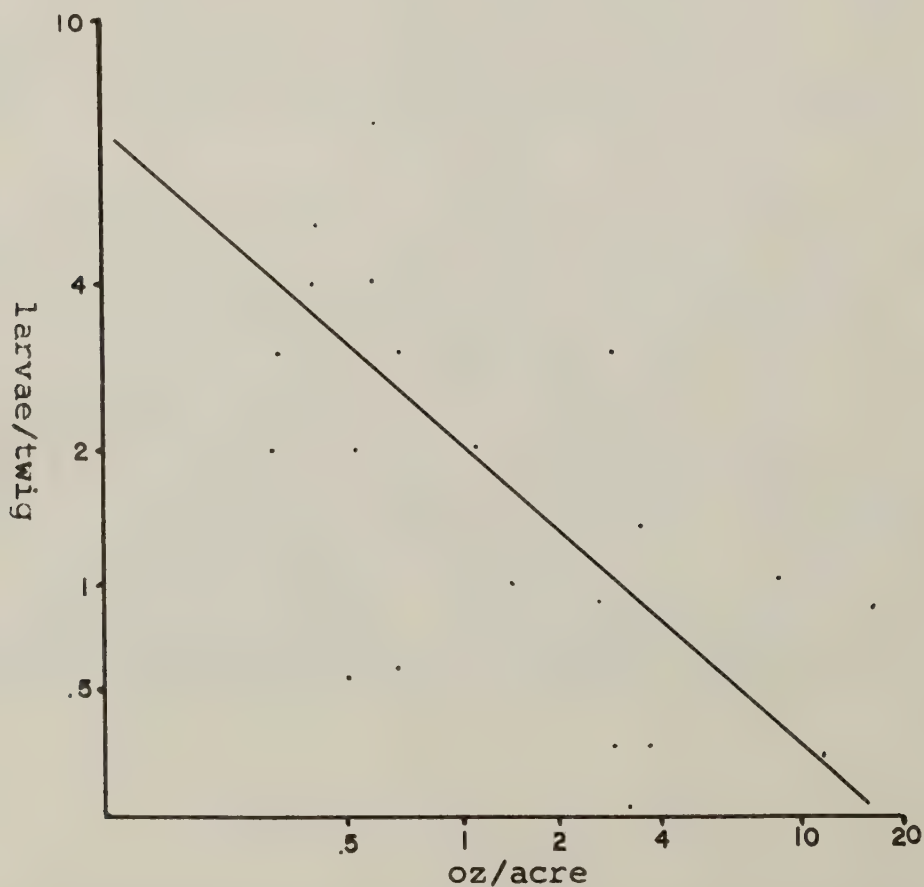


FIGURE 4. Relationship of total spray deposit (X) to numbers of spruce budworm survivors (Y).  $r = -.59$  Plots I and II.



No additional points other than the data from plot II have been entered on the graph (Fig. 6) since as the number of points increase, overlap of points increases and the suggestion of two trends becomes obscured. If the data from plot IV, all of which received substantial coverage in both applications, is added, it corresponds closely with the right hand trend. Three appropriately treated clumps from plot I fit the left hand trend.

The figure indicates that a total application of 2 ounces, all applied in the second treatment, was nearly as effective in plot II as double this amount applied as two separate treatments, in terms of reducing spruce budworm populations. Stated another way, nearly all of the budworm population reduction experienced in this plot could be accomplished with the second application of Accothion alone.

A less extreme, but additional, confirmation of this is the considerably lower value for Y-axis intercept in the regression equation for the second application and survival, Figure 3, compared to the first application and survival, Figure 1.

A major point to be made from examination of all of the graphs presented above is that, without need of extrapolating beyond the data, higher than the recommended and applied dosages of Accothion can produce lower survival levels of budworm and somewhat lower levels of defoliation than the averages experienced in this project. This is evident from an inspection of those data points on the various graphs and comparing those tree clumps that received higher dosages with those receiving an average dosage.

### Discussion

The data presented here agree well with those determined from the extensive surveys of the Maine Forest Service that covered the entire spray area. Estimates of foliage saved and of budworm population reduction for the small plots, at least those that received the recommended spray dosage, are not widely divergent from the data presented by Coughlin.

Evidence from examining functional relationships between spray deposit, survival, and defoliation confirms Canadian experience with similar treatments wherein the first application makes its largest contribution to foliage preservation while the second application contributes most to budworm population reduction.

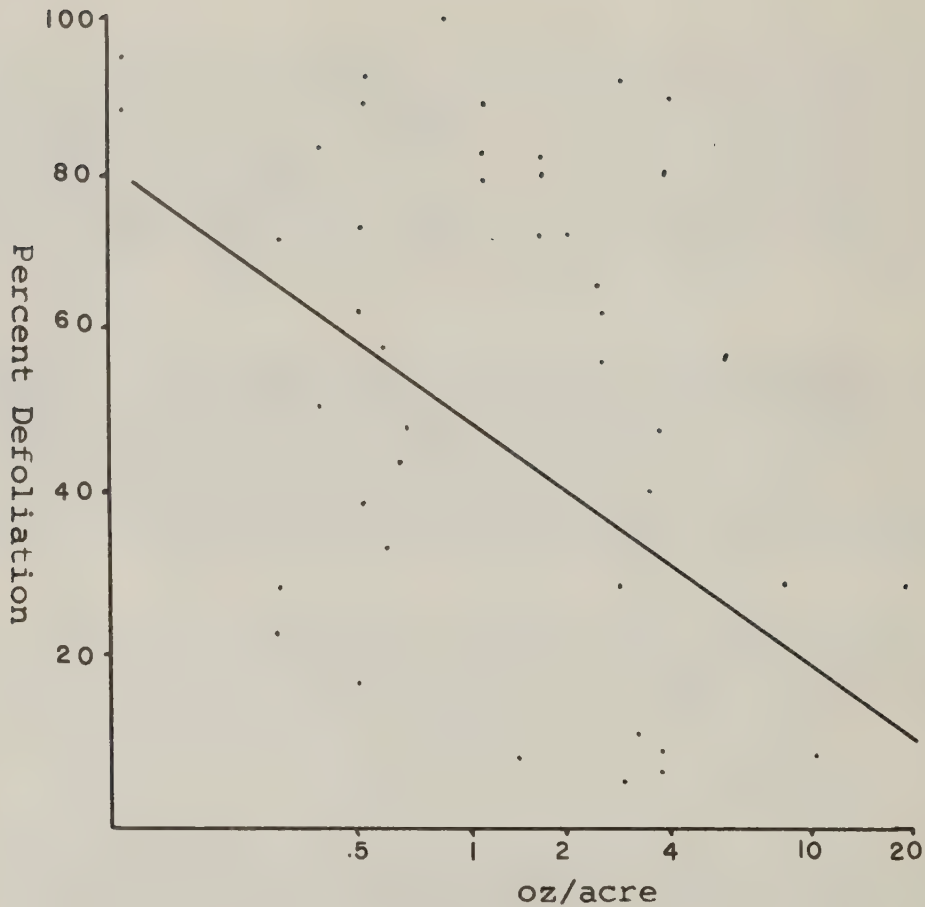


FIGURE 5. Relationship of total spray deposit (X) to percent defoliation (Y).  $r = -.47$  Plots I and V.

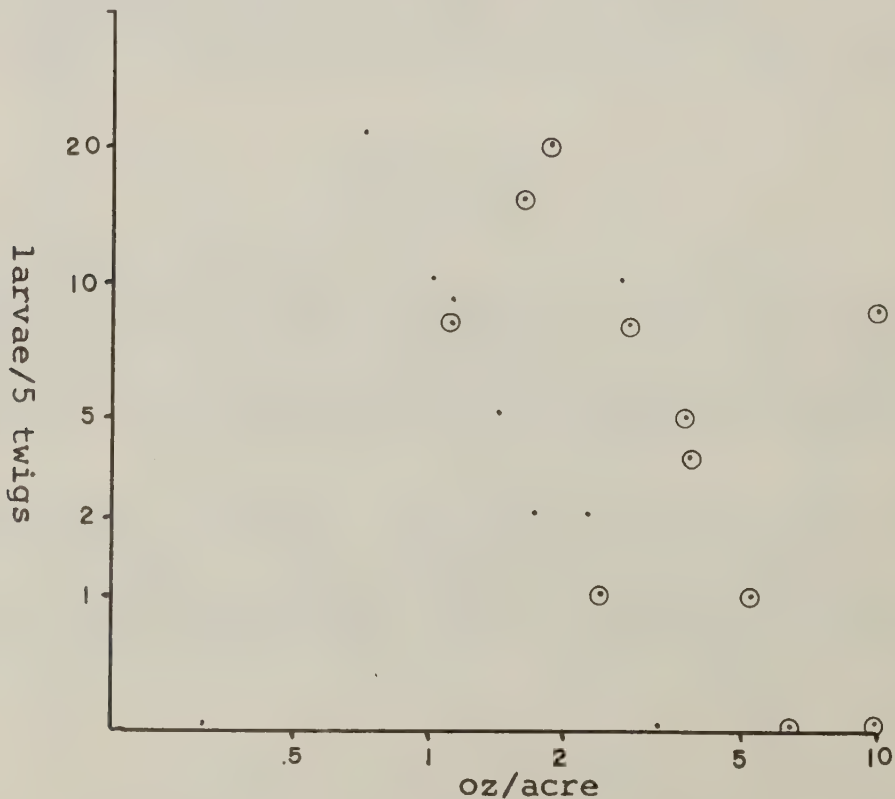


FIGURE 6. Relationship of total spray deposit (X) to numbers of spruce budworm survivors (Y) on Plot II. In this case, Y is plotted as numbers of larvae per 5 twigs.

One is able to predict from the dosage-mortality and dosage-defoliation data presented here, however, that greater foliage savings and greater population reduction can be achieved with higher dosages of the chemical. Since the evaluation of this application of Accothion has been that the level of foliage preservation was satisfactory, efforts to improve the treatment need not be concentrated on the first application, the prime function of which was foliage preservation. Rather, any future testing of higher dosages might be concentrated on the second application, which contributed the most to population reduction.

While the data suggested that higher dosages could produce better budworm control it is difficult to predict whether the improvements in control will be sufficiently pronounced to warrant the added economic and environmental expense. While the graphed trends are suggestive, precise predictions cannot be made because of the low values for the correlation coefficients, deriving from the high variability in the data. The question can only be resolved by further testing.



## Effects of Accothion on Parasites and Predators

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During the pilot test to determine the effectiveness of Accothion against the spruce budworm in Maine, data were collected to assess the effects of the insecticide on parasitic and predaceous insects. Several approaches were used, including the collection of insects poisoned by the insecticide, dissections of larval spruce budworm for parasites, and rearing of late-instar budworm larvae for parasites.

The study reported here was made in cooperation with the U.S. Forest Service. The dissections and identifications of the immature parasites were made by personnel of the Maine Forestry Department.

### Methods

Slightly more than 200,000 acres in Aroostook County, Maine were treated with two applications of 2 oz/acre of Accothion. The study area in the sprayed region consisted of five lines each having 50 numbered fir and 20 numbered spruce trees, mostly red spruce. Four of the test lines were in the town of Oxbow: line I, along Route 11, and lines II, III, and IV along the Oxbow Road. Line V was in Masardis, off the Garfield Road near Rockwell School (See map in Appendix). Two check areas were established outside the sprayed area, one in Masardis near Blackwater Stream (Blackwater check), and the other in Garfield and T11 R7 along the American Reality Road (Reality check). The study areas are the same as those discussed by Chansler and Dimond elsewhere in this report.

The spray dates for the first application of Accothion on all lines was June 4. The dates for the second application were June 10 for lines I and II; June 14 for lines III and IV.

To determine the kinds of insects killed by Accothion, cotton muslin cloth was cut into 6x6 ft squares and placed under each of 5 fir trees along lines I, II, III, and IV prior to the first application. The cloths were stapled to pegs placed along the edges. The edges were elevated about 6 inches so insects landing on the cloths would remain. The cloths were checked the day prior to each application of Accothion and any debris and insects on them were removed. The cloths were again checked approximately 24 and 72-hours after each spray. All insects and spiders on the cloths were collected and preserved in 70% alcohol with the exception of some of the live spruce budworm larvae collected 72-hours after the second application of Accothion. These larvae were placed in petri dishes with the synthetic food of Leonard and Doane (1966) to determine the residual effectiveness of the insecticide.

Many of the larvae in the early collections of foliage were preserved in alcohol and dissected for parasites.

Many of the larger larvae which were alive when collected on foliage after the second application of Accothion were placed with fresh foliage to be reared for emergence of parasites. Within several days, these larvae were placed in petri dishes with synthetic food and brought to the University of Maine insectary at Orono. These larvae were kept in a controlled room at 23.5°C with synthetic food. In August, after development was completed, the number and kinds of parasites in each dish were recorded, along with the number of spruce budworms that had developed into adults.

## Results

### Drop-cloth collections: parasites

The numbers of parasitic insects collected on the 20 drop-cloths after applications of Accothion are given in Table 1. A total of 65 specimens of parasites were recovered. Most of the parasites were collected after the first application of the insecticide. Of these, 30 were parasitic wasps

Table 1. Numbers of parasitic Hymenoptera and Diptera collected on drop-cloths after applications of Accothion

Line	Tree	<u>1st Application</u>				<u>2nd Application</u>			
		<u>24 hr</u>		<u>72 hr</u>		<u>24 hr</u>		<u>72 hr</u>	
		Hymen.	Dipt.	Hymen.	Dipt.	Hymen.	Dipt.	Hymen	Dipt.
I	2	1	0	1	0	0	0	0	0
	8	4*		0		2		1	
	19	0		2		3		0	
	21	0		0		0		0	
	41	1		0		1		0	
II	1	2	0	0	0	0	0	0	0
	8	3		0		0			
	32	1		0		0			
	47	3		1		5*			
	48	2		0		0			
III	9	3	0	0	0	0	1	0	0
	13	1		0		2	2	0	
	33	1*		1		0	0	1	
	35	1		0		0	0	1	
	43	2		0		0	0	1	
IV	8	0	0	2	0	3**	0	0	0
	34	1		0		0			0
	39	3		3		0			0
	41	1		1		0			0
	43	0		0		1			1
		<u>30</u>	<u>0</u>	<u>11</u>	<u>0</u>	<u>17</u>	<u>3</u>	<u>4</u>	<u>1</u>

\* Includes 1 Meteorus trachynotus (Vier.)

\*\* Includes 2 Glypta fumiferanae (Vier.)



and 10 were dipterans (tachinids). Only two specimens were recognized as spruce budworm parasites, both of them identified as Meteorus trachynotus (Vier.), a braconid that attacks late-instar budworm larvae. Diagnosis of the tachinids upon recovery from alcohol proved difficult, and they were retained to be shipped off for identification. This collection may include budworm parasites.

The numbers of parasites collected after the second application of Accothion were fewer; about one-half as many as were found on cloths after the first application. Most were parasitic Hymenoptera. Two species of budworm parasites were recovered; M. trachynotus and Glypta fumiferanae (Vier.). The latter parasite occurs in early larval stages of budworm, and those killed were ones which had completed their development and had emerged from budworm larvae.

Most of the parasites were collected in the 24-hour samples. Only 5 of 65 parasites were found in the 72-hour samples, all after the second application of Accothion.

Common in the drop-cloths, but not identified, were spiders. Spiders are the most important arthropod predators of spruce budworm larvae (Morris 1963a).

#### Drop-cloth collections: non-target insects

The broad insecticidal properties of Accothion are evident from the numbers and the diverse taxa of non-target organisms collected on the drop-cloths. Nematocerous dipterans were the most numerous. At some sites, plecopterans were collected in large numbers. Coleopterans were found in the following order of frequency: coccinellids, particularly Mulsantia hudsonica (Casey), elaterids, scarabids, and lampyrids. Among the hymenopterans other than parasitic wasps were formidids and several tenthridinids. The Lepidoptera were predominantly spruce budworm larvae, but several other tortricids and geometrid larvae were also found. Of Heteroptera, there were several pentatomids and a lygaeid. The homopterans included several aphids. Along with the nematocerous dipterans and plecopterans, several other aquatic groups were represented, including ephemopterans and trichopterans.

All of the specimens collected on the drop-cloths will be

retained in the University of Maine collection at Orono for at least 5 years, and will be available to anyone wishing to view them.

Drop-cloth collections: spruce budworm larvae

The numbers of spruce budworm larvae collected on drop-cloths after each application of Accothion are shown in Table 2. From 20 drop-cloths, representing a maximum of 720 square feet, a total of 5767 budworm larvae were recovered. The mean number of larvae per cloth for the total of 4 collections was 288. The lowest number was 40 under tree 34, line IV. The highest number was 663, under tree 44, line IV.

Of the total larvae recovered, only 15% were obtained after the first application. However, these data could be somewhat misleading for the following reasons:

(1) Line I did not receive a full dosage of insecticide in the first application (Chansler and Dimond, this report). Some of the larvae killed by the second treatment on this line might have been killed by the first application of Accothion had it been applied at the proper dosage.

(2) At the time of the first application (June 4) some larvae were still mining needles, and these larvae, if affected by the insecticide, would not be represented in the drop-cloth collections.

(3) Accothion is apparently a very virulent poison against spruce budworm larvae. Poisoned larvae discharge a dark liquid from both the mouth and anus. This discharge is sticky, and many larvae were glued to the drop cloth. Undoubtedly, larvae were also stuck to the foliage, and did not fall to the ground. When the second application of Accothion was applied, the budworm larvae were larger. Although these larvae reacted to the insecticide in the same way, their increased weight may have reduced the numbers adhering to the foliage. Fewer dead larvae were recorded from the foliage after the second application of Accothion.

The data from the drop-cloths indicate that Accothion does have some residual effectiveness. Large numbers of larvae were recovered 24 to 72 hours after treatment. With few exceptions, all larvae on the cloths were dead 72 hours after the first application. However, 72 hours after the

Table 2. Numbers of spruce budworm larvae collected on 20 drop-cloths after applications of Accothion

Line	Tree	1st Application		2nd Application	
		24 hr	72 hr	24 hr	72 hr
I	2	0	0	34	65
	8	1	0	487	96
	19	12	14	187	94
	21	13	16	127	26
	41	36	73	214	28
II	1	3	9	26	18
	8	26	12	114	76
	32	56	61	14	0
	47	101	26	185	8
	48	5	3	306	42
III	9	28	15	53	94
	13	40	29	101	108
	33	19	0	273	349
	35	44	12	152	160
	43	7	0	17	64
IV	8	11	12	73	72
	34	3	0	16	21
	39	12	23	262	103
	41	5	0	181	131
	44	76	91	308	188
Totals		498	396	3130	1743



second application a number of budworm larvae were alive. Some of these survivors were placed in petri dishes with artificial food. Survival of these larvae can be seen in Table 3. Of the 377 larvae collected on 8 cloths, 86 (22.8%) were alive. On June 18, eight days after treatment, the number alive was 37 (9.8% of the total collection and 43% of the number alive on June 13.) Four (10.8%) of the survivors were parasitized and one larva died of unknown cause. The remaining 32 larvae survived to adulthood.

#### Parasite survey: larval dissections.

Some of the spruce budworm larvae in the foliage collections made by the U.S. Forest Service and Maine Forestry Department, were dissected for parasites. The parasites surveyed by this technique included Apanteles, Horogenes, and Glypta fumiferanae. The numbers of Horogenes were very small, and are not included in this report.

There are a number of species of Apanteles that attack spruce budworm larvae. According to Miller (1963b) A. fumiferanae is the most common species, and the data presented here most likely represent this species.

The data from more than 5100 dissections are presented in Table 4. In the pre-spray samples, collected on June 27 and 28, 3320 larvae were dissected, including 199 from Blackwater check. In the post-spray samples, taken on July 8 and 9, 1800 larvae were dissected, including 183 from Blackwater check and 225 from Reality check.

A comparison of the percentage of Apanteles found in the pre- and post-spray samples show almost a 3-fold increase in Apanteles in the post-spray samples. However, the Blackwater check samples show a similar increase, indicating that the increase in parasitism is not due to the application of Accothion but rather to the ability of those making the dissections to distinguish parasites more readily in the larger larvae. This is understandable, for the larger budworm larvae are easier to dissect and the parasites are larger and more readily observed.

Although the pre-spray collections indicate a low amount of parasitism, the percentages of parasitism for both Apanteles and Glypta are rather uniform, with plot III showing the highest percentages. For Apanteles, the lowest percentage, in line IV, also is the lowest in the post-spray collection for that line.

Table 3. Survival of spruce budworm larvae collected alive on 20 drop-cloths 72 hrs after the second application (June 10) of Accothion

Line	Tree	No. Larvae June 13	No. Alive June 13	No. Alive June 17
I	2	65	17	6
	8	96	6	4
	19	94	15	4
	21	26	17	11
	41	28	6	1
II	1	18	9	4
	47	8	5	1
	48	42	11	6
		<u>377</u>	<u>86</u>	<u>37</u>

Table 4. Results of dissections of budworm larvae for Apanteles and Glypta. The post-spray collections of budworms were made after the 1st application of Accothion.

<u>Pre-spray collections</u>					<u>Post-spray collections</u>			
Line	Date of Colln.	No. diss.	% Apan.	% Glyp.	Date of Colln.	No. diss.	% Apan.	% Glyp.
I	V-27	740	5.14	2.43	----			
II	V-28	809	6.30	2.60	----			
III	V-28	743	10.90	4.58	VI-8	674	27.45	3.41
IV	V-28	829	4.58	3.26	VI-8	726	11.98	4.68
Black-Water	V-27	199	8.04	2.51	VI-9	183	21.86	4.92
Reality Check					VI-9	225	23.11	3.56

From these data it is not possible to determine whether Accothion has any effect on the percentage of parasitism of survivors of the treatment.

#### Differences in parasitism in balsam fir and spruce

Foliage samples from each tree were kept separately. This made possible comparisons of percent parasitism of Apanteles and Glypta in fir and spruce. These data are presented in Table 5. Data for line V are omitted because the foliage collections were made on a number of dates.

The data for spruce presented for lines I, II, and IV come from red spruce. The larvae in line II were collected on black spruce (60.1%), red spruce (33.1%), and white spruce (6.8%).

Data from every sample show that the percentage of parasitism in balsam fir was higher for both Apanteles and Glypta. In the collections taken on June 27 and 28, a total of 11.6% of the budworm larvae were parasitized by Apanteles on fir, compared with 6.7% on spruce. With G. fumiferanae the corresponding percentages are 3.9 and 1.9. In the June 8 collections, the total parasitism of Apanteles on fir was 18.1%, and on spruce, 13.9%. For G. fumiferanae, 4.3% of the budworm larvae were parasitized on fir, and 2.3% on spruce. In all, the ratio of parasitism of fir/spruce is 1.74 for Apanteles, and 2.05 for G. fumiferanae.

#### Parasite survey: rearing of budworm larvae

Live budworms on the foliage collected after the second application of Accothion were placed with synthetic food for rearing. A total of 3173 budworms were obtained from lines II, III, IV, V, and Blackwater check. About 7 percent of the total sample was in the pupal stage at the time of collection, with the highest percentage, 22%, in line II.

The data on the percentage parasitism in late-instar larvae are seen in Table 6. No data are given for line I, as these larvae were not kept for rearing. The average emergence of budworm adults in all collections was 63.2%.

The numbers of Apanteles are low, for most had emerged before the collections were made. G. fumiferanae were beginning to emerge at the time the larvae were collected and the percentages obtained were slightly lower than those in the post-spray dissections listed in Table 4.



Table 5. Differences in spruce budworm parasitism on balsam fir and species of spruce. Data for spruce on lines I, II, and IV were all from red spruce, and on line III a mixture of species, with black spruce predominating.

Line	Date of Colln.	No. larvae dissected		% Apanteles		% Glypta	
		spruce	fir	spruce	fir	spruce	fir
I	V-27	223	516	4.04	5.62	1.80	2.71
II	V-28	165	644	4.85	6.68	1.21	2.95
III	V-28	148	743	8.78	10.90	4.05	4.58
IV	V-28	245	584	3.67	4.97	4.00	4.79
III	VI-8	73	601	26.02	27.62	0	3.83
IV	VI-8	100	626	5.00	13.10	4.00	4.79

Table 6. Results of rearing budworm larvae for parasites

Line	Date of Colln.	Total Colln.	No. of Larvae	% emergence adult	% Apan.	% Glyp.	% Meteor.	% Horo.	% Dipt.
II	VI-17	396	392	57.1	1.79	2.55	0	0	0
III	VI-23	508	396	67.7	1.01	3.03	0.51	0	0
IV	VI-23	339	312	62.5	0.64	2.56	0	0	0.64
V	VI-18	1621	1544	62.4	0.65	4.47	0.13	0.06	0.06
Check Black-water	VI-16	309	303	68.1	0	3.63	0	0	0.33

The numbers of other species of parasites are all quite low, in the sprayed areas and in the check area.

Larvae collected in the Maine Forestry Department plots were also reared for parasites. The collection dates of these samples varied, however, making direct comparisons among all samples difficult. Since a different parasite fauna would be expected with the collections containing large numbers of pupae, the data were arranged according to the percentage of budworm pupae in the collections. These data are shown in Table 7. The only comparison between Accothion treated and untreated areas is in the collections containing 0 to 10% pupae. Here, some differences in parasitism between the sprayed and unsprayed region can be seen. These data suggest that the percentage of parasitism of G. fumiferanae is higher in the treated area, and those parasites not in budworm larvae at the time of application of Accothion are reduced in number. More data are needed, however, to determine if these differences are real, or caused by sampling error.

### Conclusions

Accothion can affect spruce budworm parasites and predators either directly by killing those exposed or indirectly by killing the host budworms.

Dead parasitic wasps, tachinids, pentatomids, and spiders were recovered in drop-cloths after Accothion was applied. Two species of parasitic wasps were identified in drop-cloths; Meteorus trachynotus and Glypta fumiferanae. M. trachynotus parasitizes late-instar budworm, and the G. fumiferanae had emerged from budworm larvae early enough to be killed by the second application of the insecticide. Identification of the tachinids collected on the drop-cloths may increase the numbers of budworm parasites known to be affected at the time of application.

In a study such as this, it would be useful to know what parasites are active when insecticides are applied, and what their population densities are. For most of the budworm parasites, the time of activity in spring is not yet known. The density of the parasites is difficult to assess, except by comparison of the percentage of parasitism in sprayed and unsprayed areas.

The percentages and parasitism by species attacking late-instar and pupal budworms were low. This may indicate a

Table 7. Results of rearing spruce budworms from the Maine Forest Service plots. These collections were made over a period of time and the collections are arranged according to the percentage of pupae in the collections.

Treatment	No. of Larvae	% pupae	% Glypta	% Meterous	% Horogenes	% Ito- plectes	% Dipt.
Check plots	172	0 to 10	4.65	2.91	0	2.33	4.07
Accothion	201	0 to 10	7.96	0	0	1.00	1.00
Accothion	378	11 to 50	4.23	0.79	0.26	2.12	0.79
Accothion	406	51 to 80	1.97	0.25	0.25	1.97	1.97



Cotton muslin drop-cloths were used to catch insects falling from trees sprayed with Accothion.



low population density for each of the species that attack these stages, but in the study plots, few collections were made of later stages of budworms, and none approached a collection containing 50% pupae.

In some instances, the application of insecticides has been shown to increase the percentage of parasitism of species in the host at the time of application. Although the collections of budworm larvae on the test lines should have shown whether such an effect occurred with Accothion, differences in the ability to differentiate parasites in dissections of smaller budworm larvae make comparison of pre- and post-spray samples difficult. Data collected from rearing larvae from Maine Forestry Department collections suggest that there might be such a relationship for G. fumiferanae. However, the difference is not large, and one might argue that the loss of numbers in budworm larvae killed by the insecticide, and the large numbers of budworms that survive to the next generation could result in a reduction of the percentage of parasitism in the succeeding year.

As far as I know, the difference in percentage parasitism of budworm larvae on fir and spruce has not previously been recorded. In an epidemic population, like the one studied, this differential in parasitism probably has little significance, but in endemic populations, it could be an important factor, particularly since outbreaks usually occur in stands of fir (Greenbank 1963). The higher incidence of parasitism of Apanteles and G. fumiferanae may help delay the release of an endemic population.

It has proven to be difficult to obtain sufficient data on parasites following the collecting regime of this test designed primarily to determine budworm mortality. More definitive data on parasites could have been obtained with collections taken specifically to survey late-larval and pupal parasites. Also, more check collections would have been useful for a better comparison of treated and untreated areas.

More life-history information of the parasites of the budworm is needed to know which ones are active at the time of insecticide application. In future spraying operations, valuable data could be obtained with more extensive use of drop-cloths to assess what parasites are killed by the spray.

#### Acknowledgements

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dissection data, large larvae for rearing, and the loan of a reference collection of budworm parasites, I extend my thanks to Mr. John Coughlin, Maine Forestry Department. For assistance with rearing of the budworm larvae, I thank Mr. Jere Downing.

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Accothion and Aquatic Insects:  
Monitoring of Stream Populations\*

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Stream-invertebrates, chiefly insects, have often been used to monitor effects of pesticide applications. They appear to be sensitive to rather low dosages of many insecticides, and population changes are rather easily measured. Existing sampling techniques, however, are subject to wide sample variation so that only major population changes can be detected. Because of this, it has been suggested that two independently-measured types of samples be taken concurrently to increase confidence in the results (Dimond 1967). The two methods generally employed are bottom sampling (Surber 1937) and drift analysis (Muller 1954).

Both methods were employed on several streams sprayed with Accothion in 1970, and concurrently on unsprayed streams for comparative purposes. The results of that study are presented below.

Methods

Twelve streams were studied, six of which were completely encompassed within the Accothion spray area. Several of the sprayed brooks had a history of DDT-treatment, in 1967 or earlier years. In each case, however, these were matched with streams outside of the Accothion area with similar DDT-treatment history.

In both sprayed and unsprayed categories, four streams were selected for both bottom and drift sampling, while drift sampling alone was employed on two. Stream identities and

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types of samples taken are presented in Table 1. (See Map in Appendix).

Samples were taken on three occasions, 1-4 days before any treatment and 3-4 days following each of the two applications of Accothion. A fourth sampling period in late summer was contemplated but abandoned when analysis of the earlier samples showed only minor apparent effects of the spray.

At each sample period, collections were made in the following manner: Bottom samples were taken with conventional, square foot Surber sampler. This device captures in a downstream net the organisms dislodged from prolonged agitation and stirring of a square foot of stream bottom. Nine replicates, three from each of three successive riffles, comprised a sample from each stream and date. The replicates were pooled producing a single sample representing the invertebrate fauna of nine square feet of bottom. The samples were preserved with 80% ethyl alcohol and later sorted, counted and identified in the laboratory. Bottom samples were taken in the same riffles on each sampling date. Different parts of the riffles were sampled each time, however, to avoid multiple collection of the same substrate. Drift samples were taken by placing one net of square foot opening in each stream for 24 hours. Samples were preserved and sorted as indicated above.

### Results

The general results of this study are illustrated in Table 2 which presents the treatment totals. Bottom samples showed no difference between sprayed and unsprayed total organisms except in the 2nd postspray collection. The difference, which is not great, was reflected in both numbers and volumes of insects. This effect was not noted in total drift of organisms, however, where there were essentially no differences in numbers between sprayed and unsprayed streams. There was a decline in volume of drift indicating that the numbers involved small species.

It should be noted here that the organisms captured in drift were living. Drift samples were delayed sufficiently long after application that floating dead organisms were flushed from the streams and not collected.

The differences noted in Table 2 are not sufficiently great to provide confidence that they represent real differences.

Table 1. Identities and locations of study streams, with description of types of samples taken.

<u>Sprayed Streams</u>	<u>Unsprayed Streams</u>
Umcolcus Stream Oxbow, B, D <sup>1</sup>	Blackwater River, Masardis, B,D
Houlton Brook, T9R6, B, D	Trout Brook, Garfield, D
Mack Brook, T8R5, B, D	Harper Brook, St. Croix, B, D
Tracy Brook, T7R5, B, D	Hastings Brook, Merrill, B, D
Shields Brook, T10R6, D	Unnamed Brook, Ashland, B,D
Otter Brook, T10R6, D	Alder Brook, Ashland, D

1 B - bottom samples  
D - drift samples

Table 2. Total numbers and volumes of invertebrates collected in sprayed and unsprayed streams. Volumes are listed in parentheses as ml. of water displaced.

	<u>Bottom Samples</u>		<u>Drift Samples</u>	
<u>Sample Period</u>	<u>Sprayed</u>	<u>Unsprayed</u>	<u>Sprayed</u>	<u>Unsprayed</u>
Prespray	2847 (20)	2992 (29)	863 (17)	649 (7)
1st Postspray	2180 (18)	1947 (16)	909 ( 6)	1109 (7)
2nd Postspray	2288 (16)	3972 (29)	691 ( 3)	488 (6)
<hr/>				
	F treatments	= .56	F treatments	= .07
	F sample periods	= .84	F sample periods	= .75

Table 3. Total numbers of baetid mayflies collected in sprayed and unsprayed streams.

	<u>Bottom Samples</u>		<u>Drift Samples</u>	
<u>Sample Period</u>	<u>Sprayed</u>	<u>Unsprayed</u>	<u>Sprayed</u>	<u>Unsprayed</u>
Prespray	1082	622	319	128
1st Postspray	833	454	255	88
2nd Postspray	598	887	70	98
<hr/>				
	F treatments	=.48	F treatments	=.85
	F sample periods	=.65	F sample periods	=2.30

For this reason, variance within and between treatments was analyzed looking for statistical significance. All calculated F values were less than 1 indicating lack of significant differences. The differences noted could quite possibly have been the result of sampling error.

As a second step in the evaluation of the collection data, the more common insect groups were examined individually. Table 3 presents treatment totals for mayflies of the family Baetidae, the most common of the Ephemeroptera in the samples.

Although the 2nd postspray collections were decidedly lower in sprayed compared to unsprayed streams, the pattern of variability was such that the differences were not significant. Among the bottom samples, the low total in the 2nd postspray collection resulted from a very low count in only one of the four sprayed brooks, the other three having counts equal to those of unsprayed streams. This one brook, Houlton Brook, showed a similarly distinct pattern in the case of several taxa, and this special case is discussed later in greater detail. Among the drift samples, it is again a single brook that gives the appearance of proportionately more insects present in unsprayed than sprayed streams after the second spray application. Actually, 11 of the 12 study brooks, sprayed and unsprayed, showed very small numbers of baetid nymphs in drift in this collection period, while in one unsprayed stream, numbers were high, contributing 81 of the 98 total listed in Table 3. It is impossible to relate this difference to spray effects.

An examination of counts of the Heptageniidae, the other common family of mayflies, showed no differences among the bottom samples of sprayed and unsprayed brooks. Total counts at all sampling periods fell between 128 and 223 individuals. This group was never abundant enough in drift samples to draw conclusions, with counts ranging from 0 - 7.

The Chironomidae, non-biting midges, did show a statistically significant effect of the spray. Data are listed in Table 4. A calculated value for F for the difference between sprayed and unsprayed streams was 5.77, significant at the 5% probability level.

With the Accothion application showing an effect on population of Chironomid larvae, it followed that other nematoceros Diptera should be examined in detail. These primitive Diptera are among the most sensitive of aquatic insects to pesticide applications. Population data for black flies (Simuliidae) are presented in Table 5. In contrast to the Chironomidae, there was no evidence of an effect of the spray application. It is of interest to note that adult



Table 4. Total numbers of chironomid midges collected in sprayed and unsprayed streams.

<u>Sample Period</u>	<u>Bottom Samples</u>		<u>Drift Samples</u>	
	<u>Sprayed</u>	<u>Unsprayed</u>	<u>Sprayed</u>	<u>Unsprayed</u>
Prespray	242	322	271	65
1st Postspray	40	166	165	71
2nd Postspray	24	411	19	233

F treatments = 5.77\*  
 F sample periods = 1.62

Table 5. Total numbers of black flies collected in sprayed and unsprayed streams.

<u>Sample Period</u>	<u>Bottom Samples</u>		<u>Drift Samples</u>	
	<u>Sprayed</u>	<u>Unsprayed</u>	<u>Sprayed</u>	<u>Unsprayed</u>
Prespray	790	1111	110	249
1st Postspray	605	650	342	495
2nd Postspray	935	1130	533	266

Table 6. Total numbers of stone flies (Plecoptera) collected in sprayed and unsprayed streams.

<u>Sample Period</u>	<u>Bottom Samples</u>		<u>Drift Samples</u>	
	<u>Sprayed</u>	<u>Unsprayed</u>	<u>Sprayed</u>	<u>Unsprayed</u>
Prespray	198	175	40	119
1st Postspray	111	193	84	380
2nd Postspray	148	296	27	45

F treatments = 1.33

F treatments = .38

black flies were affected by the spray, disappearing from the spray area for a few days after application, but this effect did not extend to the aquatic stages.

Among other Diptera, Tipuloidea and Rhagionidae were usually present in small numbers in all streams, and Blepharocerciidae were common in one sprayed stream, Umcolcus. None of these showed any reduction that could be attributed to the spray applications.

The most abundant family of stoneflies was the Nemouridae. Data for the Plecoptera are presented as totals for all families, however. Analysis of variance could not detect significant differences (Table 6).

The Trichoptera were represented by several families, the most common being Hydropsychidae. Again the data, Table 7, present totals of this family plus the sporadic collections of other families. Where totals for this and some succeeding groups are for bottom samples only, it is because these groups occur uncommonly in drift. The data show no evidence of spray effects.

Treatment totals for Odonata are also presented in Table 7. The much higher total for unsprayed streams following the second spray application was all contributed by only one of the streams, suggesting that the difference is not a result of spray application.

The only Coleoptera commonly collected were Elmidae and Psephenidae. Treatment totals for these are given in Table 8. These groups also showed little evidence of spray effects. The large total for elmids in unsprayed brooks after the second application again reflects events in a single brook suggesting no relation to spray application.

A number of additional taxa were collected but in such small numbers that population trends could not be determined. These included some Crustaceans, Annelids, Arachnids as well as other insects.

The data presented thus far is quantitative, involved with numbers of individuals in samples. An additional concern involves whether a spray application affects the quality of stream fauna. Quality is used here as related to the diversity of aquatic life, and a measure of quality is the number of taxa, or different types of organisms, present in the samples. Data on numbers of taxa in the different samples are presented in Table 9. In this case, identification of organisms was not carried below the family level, and quality is therefore expressed as numbers of families

Table 7. Total numbers of caddisflies (Trichoptera) and dragonflies (Odonata) in bottom samples in sprayed and unsprayed streams.

<u>Sample Period</u>	<u>Caddisflies</u>		<u>Dragonflies</u>	
	<u>Sprayed</u>	<u>Unsprayed</u>	<u>Sprayed</u>	<u>Unsprayed</u>
Prespray	110	231	52	26
1st Postspray	153	68	26	16
2nd Postspray	180	142	28	46
F treatments = .86		F treatments = .66		

Table 8. Total number of Elmidae and Psephenidae in bottom samples in sprayed and unsprayed streams.

<u>Sample Period</u>	<u>Elmidae</u>		<u>Psephenidae</u>	
	<u>Sprayed</u>	<u>Unsprayed</u>	<u>Sprayed</u>	<u>Unsprayed</u>
Prespray	74	218	17	5
1st Postspray	66	213	23	13
2nd Postspray	85	759	42	13

Table 9. Numbers of taxa (families) of insects identified in bottom and drift samples, presenting means and ranges.

<u>Sample Period</u>	<u>Bottom Samples</u>		<u>Drift Samples</u>	
	<u>Sprayed</u>	<u>Unsprayed</u>	<u>Sprayed</u>	<u>Unsprayed</u>
Prespray	16 (14-17)	17 (14-20)	7 (4-9)	9 (3-13)
1st Postspray	14 (10-16)	14 (12-17)	8 (5-11)	8 (2-12)
2nd Postspray	15 (13-16)	16 (15-18)	7 (1-10)	7 (3-9)



present. There is little evidence of spray effects on the quality of the stream fauna.

### Discussion

The effects of Accothion on aquatic invertebrates was moderate at most with a significant difference noted only in the Chironomidae. Although of no statistical significance, lower totals in the sprayed brooks were noted for several other taxa as well. Examinations of the data for specific streams suggests that virtually all of the difference was contributed by a single brook, Houlton Brook, which gave the appearance of important spray effects. In this brook alone, large differences occurred between the pre-spray and post-spray collection in many of the taxa. Unfortunately, being a single observation, the difference is not subject to statistical verification. The possibility is suggested that Houlton Brook received greater than average coverage from the spray application, and represents an effect of higher doses of Accothion. The spray deposit recorded on deposit cards placed within 50 yards of the Houlton Brook collection area was 6.3 ounces/acres total for the two spray applications. The remaining sprayed brooks, three in the case of bottom samples and five with drift samples, showed little suggestion of spray effects other than with the Chironomidae.

Dimond (1967, p.9) compared the effects of an application of malathion with an earlier report on impact of DDT (Gorham 1961) on aquatic insects in northern Maine streams. Whereas DDT produced reductions of 60-90 percent in the major taxa, losses caused by malathion were in the range of 50-70 percent. In both cases Plecoptera were less affected than the other groups. Accothion appears to cause as great losses of Chironomidae as DDT, but otherwise appears less destructive than either of the other two chemicals.

Regarding quality of stream fauna, malathion produced no detectable reduction in numbers of taxa and in that sense resembled the results reported here with Accothion. DDT, however, had a strong effect in this regard, reducing numbers of taxa from 19 to 5 in bottom samples and 20 to 4 in drift (Dimond 1967). Using the criterion of faunal quality, therefore, the results with Accothion also appear favorable.

Vincent (1969) reported on observations similar to the ones reported in this work, involved with a 1968 application of

Sumithion<sup>R</sup> (= Accothion) in northern Maine. In that operation, a single application of 6 oz./acre was employed, a considerably higher dosage than in the 1970 applications. Vincent reports different conclusions than those of the present study. Large losses were noted among stoneflies, for example, while Diptera showed no reductions. Since only a single sprayed stream was studied by Vincent, the significance of the differences cannot be tested, and we cannot explain the discrepancy between his results and ours.

### Conclusions

Accothion, as applied to Maine Forests in 1970, produced significant reductions in populations of Chironomidae but not in other stream organisms examined. Total numbers of different taxa in streams were also unaffected. Comparisons with other studies, involving applications of malathion and DDT to forests suggested that the impact of Accothion was somewhat milder, than with the former and considerably less destructive than with the latter.

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## Biological Observations of Fauna and Flora

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### Objective

The objective was to determine, through extensive field observations, reactions to Accothion of the general fauna and flora in the area, in particular of insects not living within water.

### Methods

Observations in the period May 29 - June 16 in the spray area consisted of searching for insects and other organisms, both animal and plant, exhibiting any spray effects or no effects. The spray area was so large that searching before, during, and after each spray could be done readily by planning daily trips in relation to blocks that had been sprayed or remained to be treated. Effort was to cover diverse territory daily and search suitable sites such as bare ground spots in woods or along woods roads as well as blossoming flowers attractive to insects. Numerous water pools in depressions of woods roads were used to real advantage because of their attraction to adults of aquatic insects, and large numbers of Amphibia; also because any insects dropping from the air or trees above would be readily revealed on the water surface.

### Results

The large number of organisms observed gave a good cross section of faunal and floral forms. Numbers of different species observed were Plants 217, Insects 126, Spiders - Unknown, Birds 73, Mammals 19, Amphibians 5, and fish and snakes 2. In addition there were observed 25 families of Musci (mosses) and 8 orders of Lichens. Detailed species names are on file.



No necrotic effects were observed on vascular and non vascular plants. A wide variety of insects showed marked effects and in numbers. In single days following sprays 1000, 500, and 200 insects - part dead, part affected - were found on water pools in narrow woods roads over which the tree canopy spread. To one familiar with insect numbers amongst and on trees, this is not a large number. The variety of affected insects represented was more than expected. Diptera (midges and gnats especially, with some syrphid and bibionid flies) were the most numerous insects followed in order by Coleoptera, Hymenoptera, Lepidoptera, Hemiptera, and a very small number in other orders. Sawflies and parasitic wasps were found equally in small numbers. Black flies and mosquitoes were far less troublesome following spraying but only for 2-3 days. Many species on the area showed no effect. Some groups showed partial effects - e.g. Geometrid moths were commonly affected but the more commonly represented species were found even more abundantly resting on tree trunks and some genera of the family were found unaffected. A number of tree-frequenting spiders were found on the water pools. Insects were far more numerous at the end of the project than at the beginning. This reflects the changing and increasing emergence and general abundance as the early season advances, rather than an influence of the spray. Likewise, it does not indicate eliminations by the spray.

Birds populations were obviously high. Apparently the abundance of budworm caterpillars as a food source attracted a high bird population. There was an especially large number of evening grosbeaks throughout the period. By observation and by stomach check of automobile-killed individuals, grosbeaks were found to be feeding heavily on budworm larvae. Migration of birds continued actively throughout the period. A ruffed grouse with at least eight new-young was observed, the young exhibiting the usual ability to scamper for hiding places. No evidence was found of injury to bird eggs, young, or adults.

High numbers of tadpoles present in the many water pools exposed to the sprays remained abundant and thriving in pools during and at the end of observations. In one trapped-pool a school of minnows was active at the beginning of operations and appeared equally as active at the end.

The mammals and the few frogs, toads, salamanders, and snakes observed showed no indication of any effect of the spray.

### Discussion

The rapid succession and change in the species of insects and birds observed and the fast development of plant growth was noticeable. The first portion of the period was excessively rainy and cool while the latter portion was dry, hot, and quickly dried some of the more shallow woods pools.

### Conclusions

Except for insects, there was no observable effect of the spray on other forms of fauna and flora covered in these observations.

Insect records indicated a correlation between the abundance of forms present at the time of spraying and the numbers of insects affected.

Records indicated phytophagous insects may be more affected by Accothion than parasitic and predatory groups.

Overall findings on all forms of life observed indicate a higher dosage of Accothion could be safely tested against the budworm in Maine.

With the observable affects limited to insects, future projects could yield more quantitative studies on affected vs. non-affected numbers.

Short Term Effects of Accothion on Fish Populations  
in Northern Maine, 1970

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Two streams within the spray area and one control\* stream north of the spray area were selected by the Maine Department of Inland Fisheries and Game to evaluate the effects of aerial spraying of Accothion on fish populations.

Shields Brook, Masardis, T10 R6, Aroostook County (see map in Appendix) is a tributary of the Aroostook River. The 460-foot sample stream section is located just above the bridge on the Craigville Road. The stream banks in this section are heavily wooded with mixed coniferous growth.

Punchard Brook, T10 R6, T11 R7, Garfield, Aroostook County is a tributary to Carry Brook in the Big Machias River watershed. The 500-foot sample stream section is located just below the McKee Road. Punchard Brook is a small cold-water brook heavily shaded by cedar growth.

Greenlaw Stream, T11 R7, T12 R7, T12 R8, Garfield, Aroostook County (the control stream) is located from 1-9 miles north of the spray area and is a tributary to the Big Machias River. The sampling area is a 500-foot section of stream located by the old Pingree Road in T11 R7, just south of the American Realty Road. Greenlaw Stream is larger and less shaded than the two sprayed brooks.

Methods

Pre-spray and post-spray population estimates were made by the Petersen mark and recapture method on each sample

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\* Synomonous with unsprayed



section using electrofishing gear.

Blocking seines (drift nets) were operated at the upper and lower ends of each sample section. The nets were maintained one week prior to spraying, during spraying, and one week after the second application of Accothion, when stream flow conditions permitted. The seines were checked every day or two, and dead fish were preserved in formalin or frozen for pesticide analysis.

Brook trout (Salvelinus fontinalis) were held in live cages within the sample sections of each of the three study brooks to determine any direct mortality caused by the Accothion spray. Each section had two live cages containing 15 hatchery-reared spring yearling trout per cage. Caged fish were placed in all sample sections on the same date after pre-spray population estimates had been made. Live cages were maintained during the same period as the blocking seines -- one week after the second application of Accothion. The live cages were checked each time the seines were checked. Samples of fish from the live cages were taken from each sample section and frozen for pesticide analysis.

### Results

Pre-spray and post-spray population estimates (with their 95% confidence limits) are shown graphically for brook trout, blacknose dace (Rhinichthys atratulus) and white suckers (Catostomus commersoni) in Figures 1, 2, and 3. Because of less efficient operation of electrofishing gear caused by extremely high stream flows, wide confidence intervals were generally found on pre-spray estimates. In all of the estimates, however, the pre-spray and post-spray confidence bands show nearly a 100% overlap, indicating no significant short-term changes in the population within each sample section (Table 1).

Fish mortality (all species) from the blocking seines is shown graphically for Shields Brook (sprayed) and Greenlaw Stream (control) in Figures 4 and 5. Both the sprayed brook and control brook experienced mortalities throughout the study period. It is believed that most of the mortality was caused by high flows and spawning movements of many of the minnow species. However, a heavy mortality of common shiners (Notropis cornutus) occurred in Shields Brook on the day of the second spray application. Samples of these fish will be analyzed for pesticide residues. There was no mortality of fish in the Punchard Brook sample section.



Drift nets were operated at the upper and lower ends of each stream sample section and were checked every day or two for dead fish.



There was no mortality of spring yearling brook trout held in live cages throughout the study period. Samples of these brook trout were taken from each of the study sections at the termination of the project, and were frozen for pesticide analysis.

### Conclusions

1. No statistically significant short-term changes (95% confidence level) could be demonstrated between pre-spray and post-spray population estimates of the three study sections.
2. There was no mortality of hatchery-reared brook trout held in live cages throughout the study period.
3. Both Shields Brook (sprayed) and Greenlaw Stream (control) experienced fish mortality throughout the study period, but no definite patterns could be established. A heavy mortality of common shiners occurred in Shields Brook at the time of the second spray application and Accothion is under suspicion. A sample of these dead shiners will be analyzed for pesticide residues.
4. In addition to the sample of common shiners, 12 samples including brook trout, white suckers, creek chubs (Semotilus atromaculatus) and redbelly dace (Chrosomus eos) will be analyzed for pesticide residues.

### Recommendations

1. While the results of this investigation have not indicated any serious short-term side-effects of Accothion on fish populations, little is known about long-term effects of repeated applications of this chemical. If repeated applications are contemplated, it is imperative that the Maine Department of Inland Fisheries and Game, in cooperation with other concerned agencies, design a more intensive research project to evaluate long-term effects of Accothion on the total aquatic environment. Funding for this type of study should be the responsibility of the agency using the chemical.



T A B L E 1 POPULATION ESTIMATES FOR 1970 ACCOTHION STUDY WITH CONFIDENCE LIMITS (95%)			
BROOKS AND SPECIES	PRE-SPRAY	POST-SPRAY	
SHIELDS BROOK (SPRAYED)			
BROOK TROUT	16 ( 3-29 )	28 ( 14-42 )	
BLACKNOSE DACE	220 ( 1-676 )	315 ( 232-398 )	
WHITE SUCKER	91 ( 1-290 )	43 ( 33-53 )	
PUNCHARD BROOK (SPRAYED)			
BROOK TROUT	113 ( 1-227 )	148 ( 129-167 )	
GREENLAW STREAM (CONTROL)			
BROOK TROUT	84 ( 1-167 )	104 ( 66-142 )	
BLACKNOSE DACE	211 ( 79-343 )	204 ( 123-285 )	

2. Although we have no data to indicate a long-term adverse effect, it is the opinion of the authors that Accothion should not be registered for extensive and repeated use until long term environmental effects are determined.

3. Use of increased dosages of Accothion should be limited to small areas for experimentation only until the effects on the total environment are determined.

FIG. 1 POPULATION ESTIMATES WITH CONFIDENCE LIMITS ( 95 % )

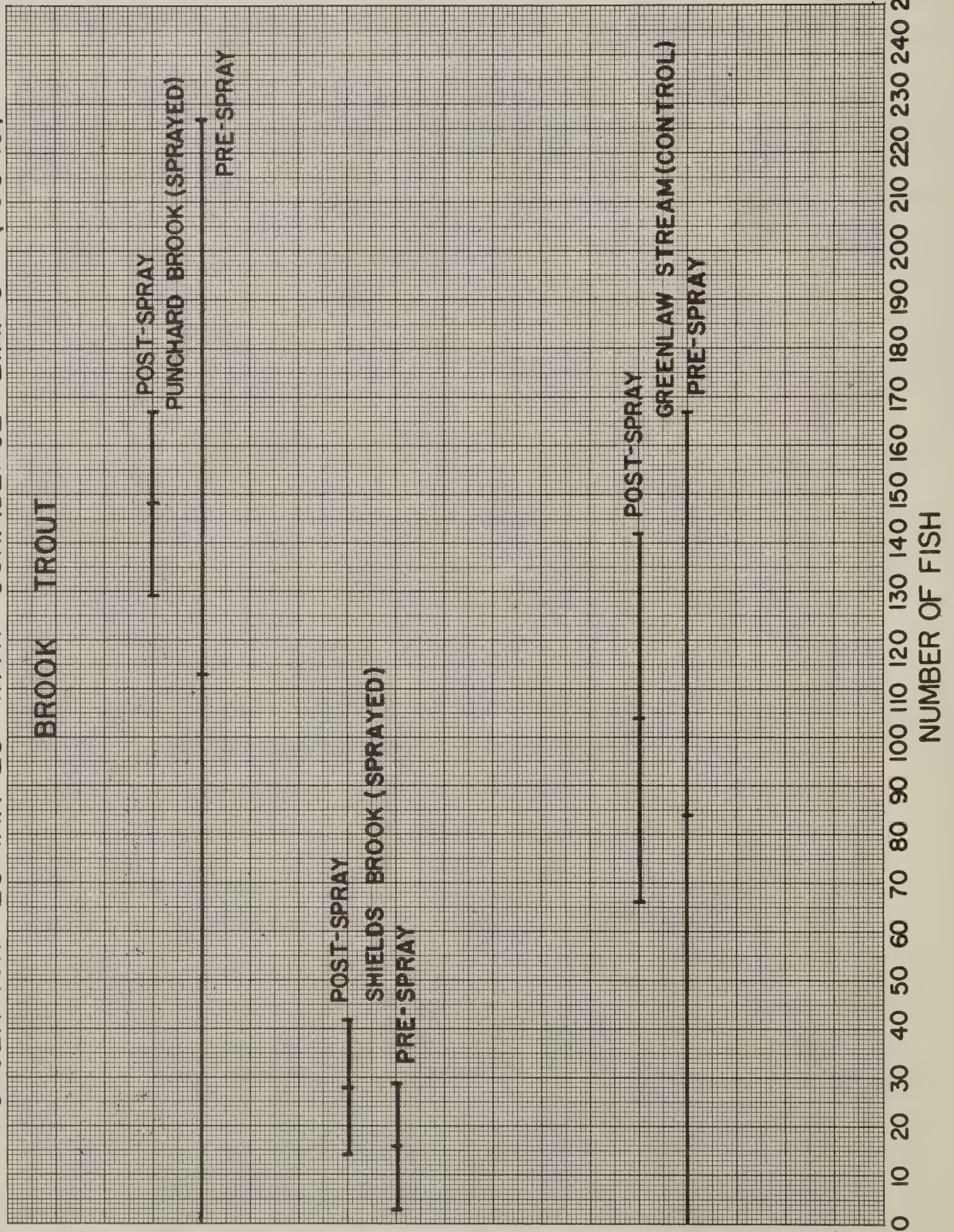




FIG. 2 POPULATION ESTIMATES WITH CONFIDENCE LIMITS (95%)

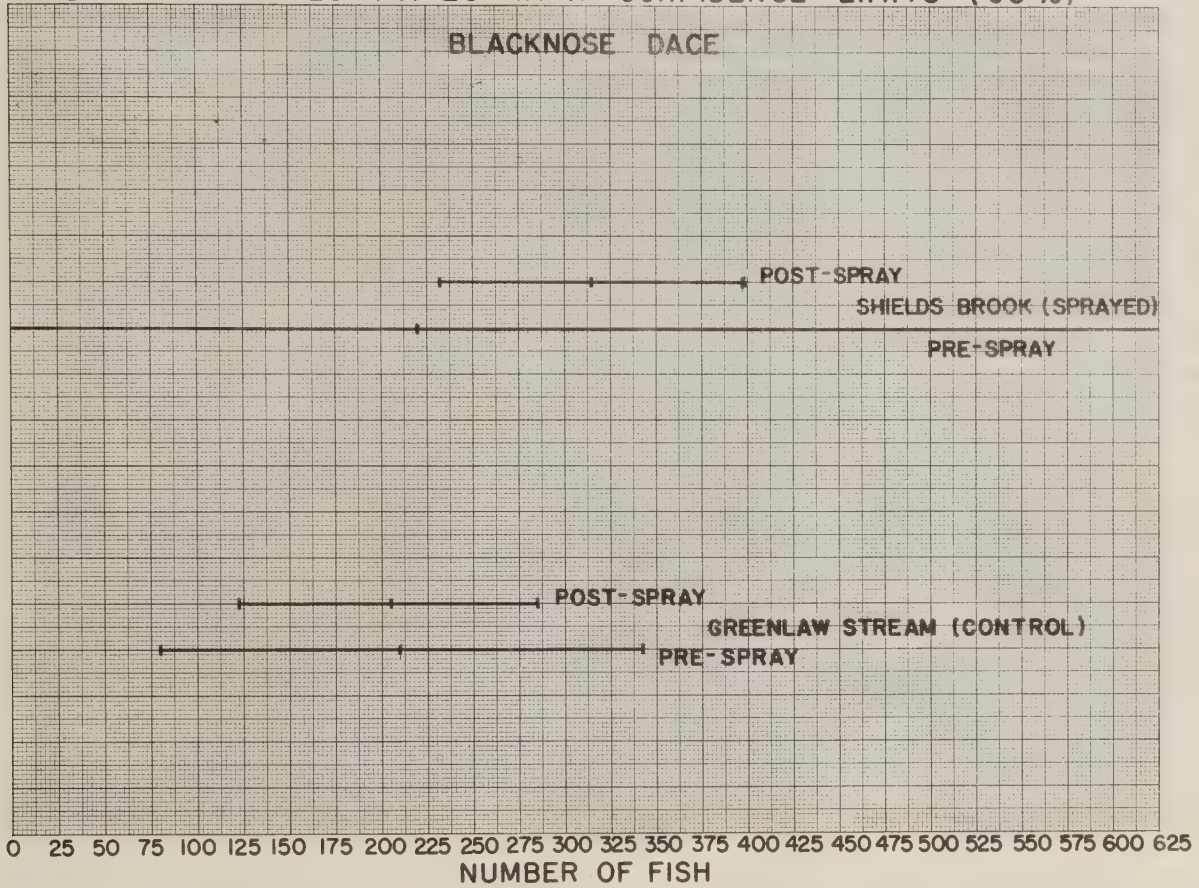
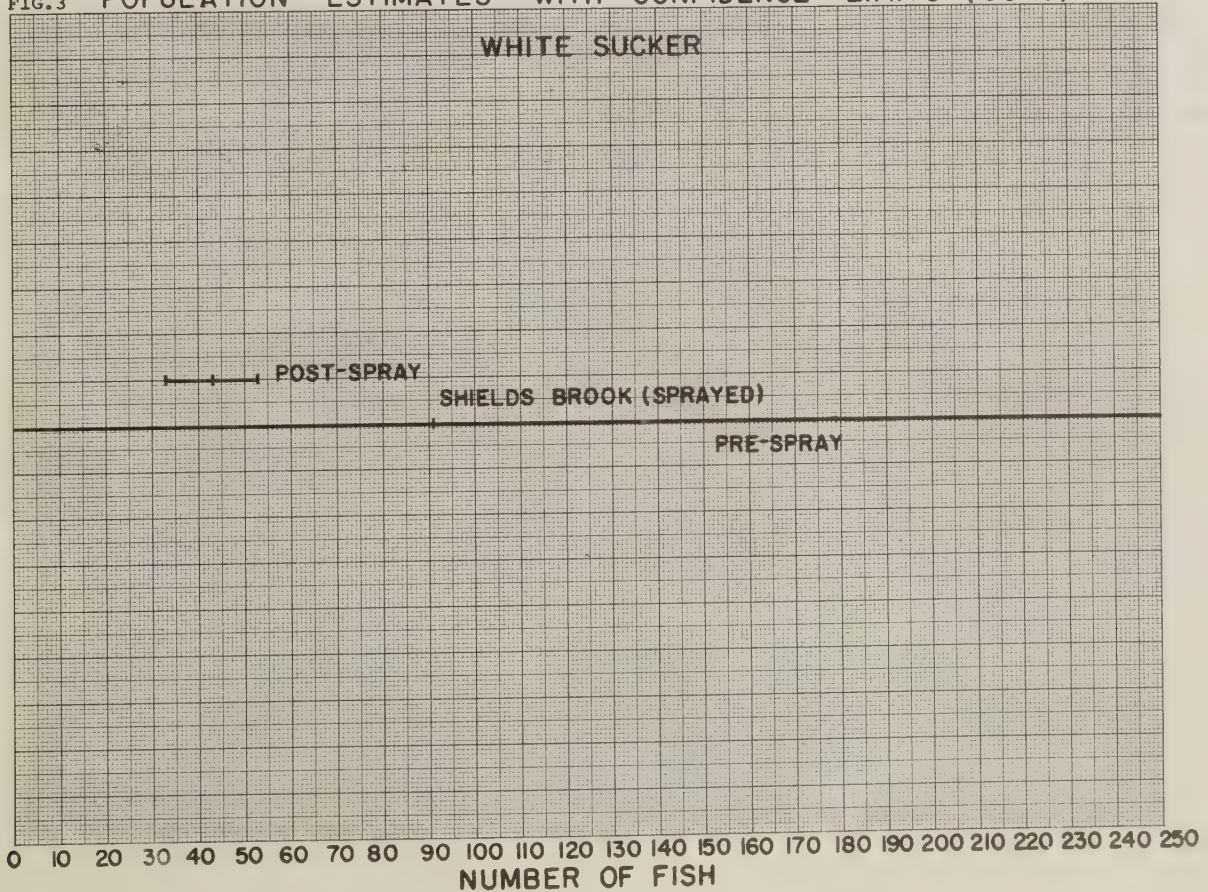
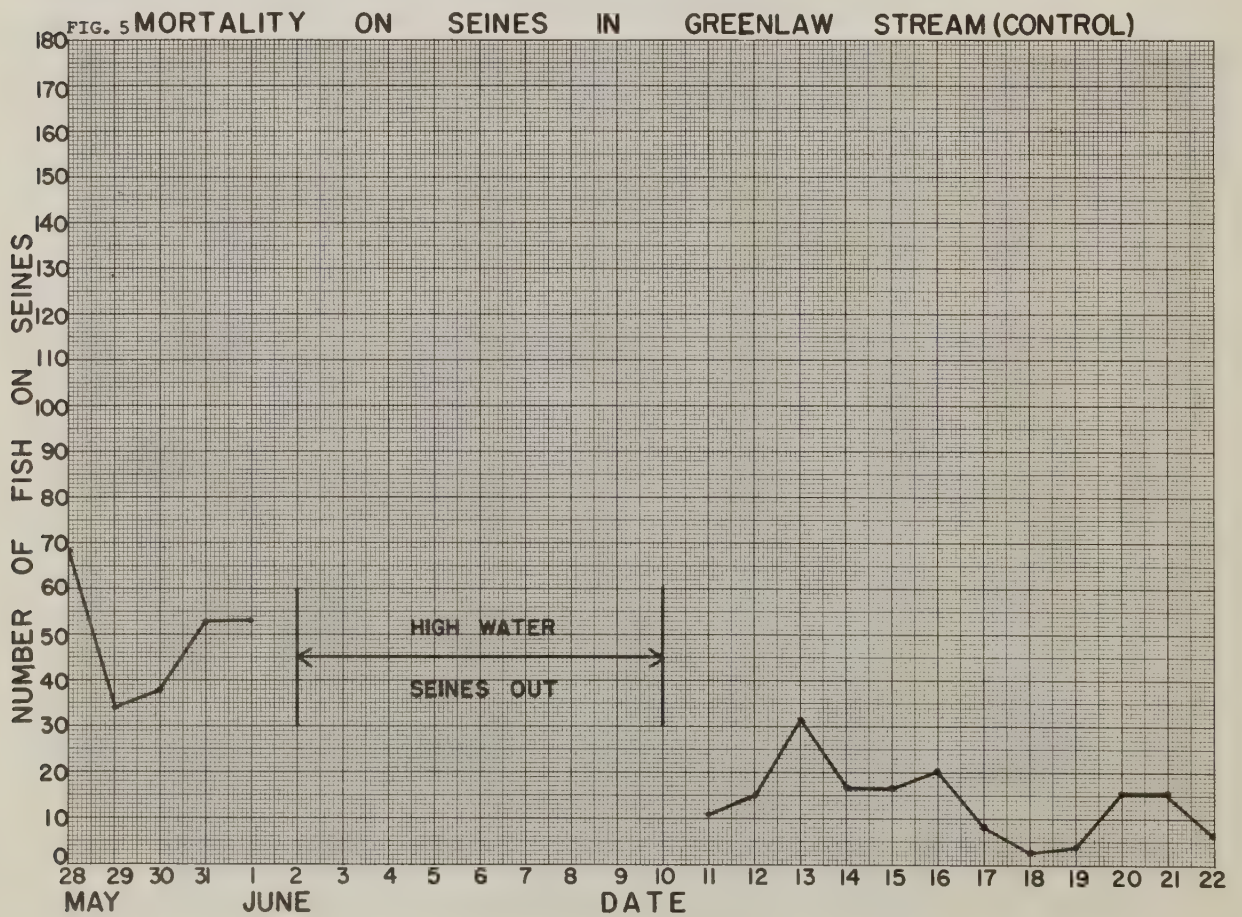
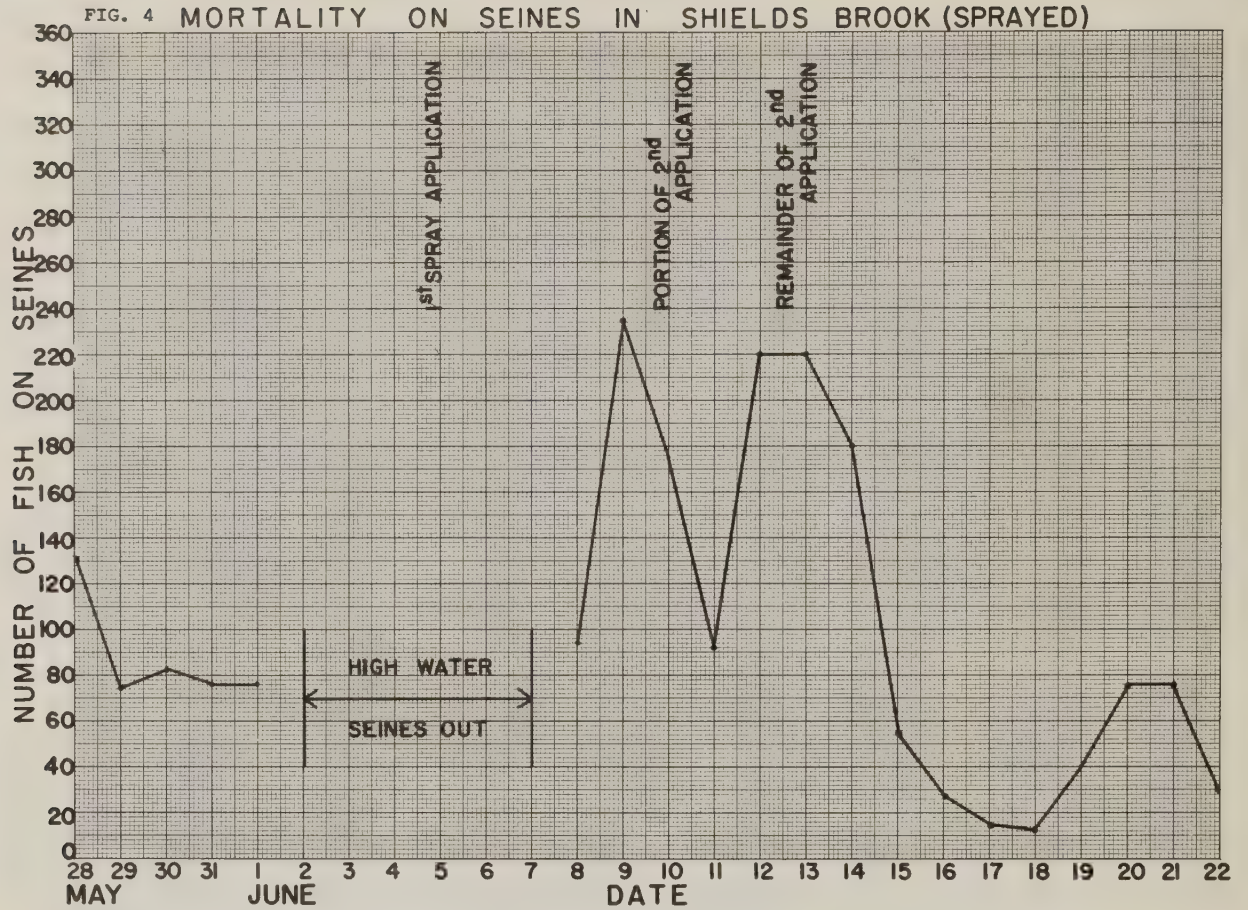


FIG. 3 POPULATION ESTIMATES WITH CONFIDENCE LIMITS (95%)









## The Effects of Accothion on Birds and other Fauna

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### Specific Objectives

- 1) To observe or detect any gross effects of Accothion on birds and mammals.
- 2) To determine sub-lethal or behavioral effects on birds by collecting brain samples for cholinesterase evaluation.

### Method

Three techniques were employed to evaluate the impact of Accothion on birds: 1) Pre- and post-spray bird censuses, 2) Carcass searches and 3) Bird brain cholinesterase determinations.

Systematic observations were made before and after spraying on six census lines; three located within the spray area and three outside that served as control lines. (See map in Appendix)\* Abandoned tote roads averaging about 1.5 miles in length were selected as census lines. Observation and listening stations were located approximately 1/8 mile apart along these lines. All six lines were walked twice prior to spraying and twice after each spray application. Each observer recorded all birds seen, or heard, at these stations during a five-minute listening period. Each census line was walked in the morning and in the afternoon to account for any differences in bird activity that could be attributed to the time of day.

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\* A control line is an unsprayed area that is used for comparative purposes.



Carcass searches and observations were made in all sprayed blocks that were accessible by road immediately following both spray applications. Personnel of the Maine Department of Inland Fisheries and Game, assisted in the carcass search. This was done by walking tote roads and making careful observations. An estimated 90 miles of old log roads were walked following the spray applications. Observations of bird activity and behavior were made. Any effects on mammals, amphibians, reptiles, and aquatic and terrestrial insects were also noted.

In addition, a Labrador Retriever was used for the carcass search. The dog was tested prior to the spray program to determine his efficiency and ability to find dead birds.

Any birds found dead or showing aberrant behavior were collected, the brains removed, and quickly frozen. Cholinesterase determinations were made later on the brain samples. This technique is specific for organo phosphate poisoning as depressed levels are indicative of an effect on the central nervous system. Regional Fish and Game personnel provided refrigeration facilities for preservation of the collected specimens.

## Results

### Bird Census

The Department of Mathematical Statistics at Rutgers University was consulted for technical assistance in data analysis. A binomial distribution treatment of the three replicates for each variable such as birds "seen" and birds "heard" indicated that the distribution was so abnormal that a standard deviation test could not be made.

A linear regression treatment was also attempted, but the time factor of the study was too short for meaningful comparison.

Although the data did not lend itself to a statistical analysis, some interesting information became apparent from the following systematic bird observations.

- 1) The number of birds and species increased from the time of the pre-census through the first and second post-censuses. This strongly suggests that migration was taking place and more birds arrived as the season progressed.

TABLE 1 ---- SPECIES COMPOSITION  
(Includes the Number of Birds Seen and Heard)

S P E C I E S*	Pre-Census		Post-Census 1		Post-Census 2		Total # of Birds	% of total
	Control	Test	Control	Test	Control	Test		
Evening Grosbeak	1	53	111	61	26	82	234	20.5
Warbler (U)	18	28	35	23	39	40	183	16.2
White-throated sparrow	35	28	34	29	27	20	173	15.3
Robin	10	16	18	14	22	31	111	9.8
Tennessee Warbler	11	13	11	19	7	30	91	8.0
Ovenbird	5	4	20	9	15	10	63	5.5
Yellow-bellied sapsucker	10	8	11	9	14	7	59	5.2
Thrush (U)	1	4	6	12	8	12	43	3.8
Nuthatch	5	3	5	6	7	6	32	2.8
Myrtle Warbler	3	1	9	7	1	7	28	2.5
Rose-breasted Grosbeak	6	6	9	3	0	1	25	2.1
Parula Warbler	0	6	2	3	4	7	22	2.0
Magnolia Warbler	7	0	4	0	5	1	17	1.5
Junco	5	2	4	1	1	1	14	1.3
Purple Finch	4	4	4	0	2	0	14	1.3
Wood Pewee	3	5	3	2	0	1	14	1.3
Swainson Thrush	2	0	4	4	0	0	10	0.9
Totals	126	180	190	202	178	256	1,133	100.0

\* (U) Indicates species unknown.

2) There was a striking difference in the number of evening grosbeaks (196) in the spray area as compared with (38) in the control area. Similar observations were reported by J. R. Blais and G. H. Parks\*, Forest Entomologists of the Department of Forestry, Ottawa, Canada. In their study of a localized budworm outbreak in Quebec, Canada, unusually large numbers of evening grosbeaks were attracted to the outbreak area during their spring migration. This influx of grosbeaks in the budworm infestation areas in both Maine and Quebec occurred when the budworm was in the late larval and pupal stages.

Table 1 shows the species composition of the spray and control areas by census. In addition to numbers of birds, percentages of the total are also presented. A perusal of table 1 shows the first seven birds in order of frequency as follows:

<u>Species</u>	<u>Total</u>	<u>Percent</u>
1. Evening grosbeak	233	20.5
2. Warbler (sp.)	183	16.2
3. W. T. Sparrow	173	15.3
4. Robin	111	9.8
5. Tennessee Warbler	91	8.0
6. Ovenbird	63	5.5
7. Y. Bellied Sapsucker	59	5.2
	<u>913</u>	<u>80.5</u>

Collectively, these seven species comprise 80 percent of the total number of birds seen and heard.

#### Carcass Search:

One dead Parula warbler, Parula americana, was found in the spray area on June 5th, the same day that this block was sprayed for the first time. Two parula warblers were collected for cholinesterase comparison; one was taken outside the spray area and the other taken inside the spray area three days after the second spray application. Results of cholinesterase tests are as follows:

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\* Blais, J. R. and Parks, G. H. Interaction of Evening Gros-Beak (Hesperiphona Vespertina) and Spruce Budworm (Choristoneura fumiferana (Clem.) in a Localized Budworm Outbreak treated with DDT in Quebec. Canadian Journal of Zoology, Vol. 42, 1964, 7 pp.



Spray bird .....	0.32 *
Control bird .....	3.72
Bird Taken in Spray area 3 days after 2nd Spray application .....	3.87

It is probable that the parula warbler with the 0.32 brain cholinesterase value was killed by the Accothion spray. Even though the sample size was too small to be significant, a comparison of the spray and control area cholinesterase values is quite suggestive.

Since there were no other dead birds found in the sample areas during the carcass search, it is presumed that no significant bird kill occurred as a result of the two Accothion spray applications. The fact that the Labrador Retriever found no birds lends further support to this assumption. There was no silence in the forest following spraying nor was there an obvious decline in bird activity or change in behavior.

Observations of other spray effect indicators were made in conjunction with the carcass searches and the bird census lines. American toads, Bufo americanus, wood frogs, Rana sylvatica, numerous green frogs, Rana clamitans, leopard frogs, Rana pipiens and pickerel frogs, Rana palustris, were seen in wheel tracks and pools of water; all behaving in a normal manner. Live polywogs were also observed in these same pools and puddles. These pools also contained living aquatic micro-crustaceans (Cladocera or Copepoda or both), and a variety of living aquatic insects (beetles including Dytiscids, and a few mosquito larval); few dead aquatic insects were seen.

Many forms of terrestrial insects were observed. Adult blackflies were very scarce in the spray area while in the control areas they were abundant and intolerable. Some adult mosquitoes were present and biting in the spray area but did not present enough of a nuisance to require the use of a repellent. Bumblebees were flying about working wild flowers in an apparently normal fashion. Small dipterans were commonly seen flying above the roads. A few of these flies were found dead on the surface of pools; these mortalities were judged to be recent, and may have been caused by the spray. Small Hymenoptera were also seen dead in some pools.

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\* Cholinesterase readings are expressed in micromoles of acetylcholine hydrolyzed by two milligrams of brain tissue in 30 minutes at 25 degrees centigrade.

During the course of these observations, a number of spruce budworms were seen hanging from upper branches of fir and spruce by silk strands. Small flocks of evening grosbeaks were observed feeding in the tops of balsam fir trees in areas where spruce budworms were spinning down on threads, but whether or not the grosbeaks were feeding on budworms is not known.

Three ground nests were observed in the spray area after spraying. The first nest contained 4 nestlings. This nest was rechecked nine days later and was empty. It is assumed that the young birds fledged as the nest itself was undisturbed and there was no evidence of predation. Two other ground nests, both with eggs were observed on this same census line. One was a White-throated sparrow nest with four eggs and the other contained seven unidentified eggs. It is not known whether these nests contained eggs during the first spray application on June 6, but the second spray on June 15 did not appear to have any effect on incubation or behavior of the parents. A White-throated sparrow nest with four eggs and incubating parents was observed in the control area after both spray applications. No difference was noted in the parental behavior of the White-throated sparrows in the spray and control areas.

### Discussion

Several variables should be pointed out relative to the pre-and post-census bird lines. For example, the pre-census was conducted on May 28 and 29, there was very little foliage on the trees and birds were easily observed. However, on June 5 and 6 when the first post-spray census was conducted and again on June 15 and 16 when the second post-spray census was carried out, foliage was quite heavy and birds were not as easily seen. It should also be recognized that migration was taking place to some degree during the pre-and both post-censuses. This means that many species of birds were transients rather than territorial.

The spray area was systematically sampled in all types of habitat with only one indication of mortality other than insects. It is the considered opinion of the investigators that any gross effects on birds or mammals caused by Accothion would have been detected.

### Conclusions and Recommendations

Based on the pre-and post-census lines and the carcass search

there was general agreement among the investigators that no significant bird or mammal kill occurred when Accothion was sprayed twice at a rate of two ounces per acre. Further, there were no indications that mammals and amphibians were affected.

Since spruce budworm control programs are usually conducted during late May and early June during the bird migration period, future bird censuses should focus on those few species that are territorial at this time.

It would be a decided advantage to know earlier whether a control program and evaluation will be conducted so that sampling designs and techniques can be planned well in advance. This would also facilitate recruiting manpower and procuring the equipment needed to carry out a meaningful surveillance program.



Effects of Accothion on Birds  
(Intensive Studies on Sapsuckers)

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Objectives

The primary objective of this monitoring was to determine whether the Accothion caused mortality or abnormal behavior among sapsuckers. A secondary objective was to explore the potentials of using sapsuckers as an indicator of the effects of pesticides on birds.

A bird species used for monitoring pesticides should permit reliable, long-term, and efficient observations, before and after application of the chemical. The yellow-bellied sapsucker (Sphyrapicus varius varius L.) appears to meet these criteria, and it has the following characteristics which make it a desirable species for monitoring pesticides in northern forests: (1) The species has highly predictable behavior patterns that assure the same birds are being observed before and after treatment. (2) Adults can be easily found, captured, and marked, or they can be observed repeatedly without capturing for at least five months (May-September). (3) In summer they are abundant in Maine and other northern forest regions. (4) They have a high degree of territorial behavior that makes them an ideal free-flying species for monitoring. (5) Adults may return to the same territory in subsequent years. (6) Sapsuckers respond readily to imitations of their drumming, so their territories, nests, and long-term feeding sites can be easily found and used for repeated observations. (7) Their food, tree sap and many insects, will be exposed to the chemicals sprayed-- as will the food of other birds.

## Methods

### Survey for Sapsucker Territories and Nests

Sapsucker territories were found by using calling - surveying techniques developed here since 1964. Two survey lines were used. Each line had 20 stations spaced 400 feet apart (1.44 miles, Fig. 1) In the spray area the line was along the Oxbow Road, between Houlton Brook and Trout Brook, in spray block 12. (See map in Appendix). It was surveyed on May 20 and included stations 1 to 20. In the unsprayed area the line was west of Ashland, along the Realty Road, between Fourmile Brook and Sixmile Brook. It was surveyed on May 26 and 27 and included stations 21 to 40.

For the survey, sapsucker drumming was imitated for at least eight minutes at each station. When sapsuckers responded by drumming, calling, or approaching, their positions and the time were plotted on the map for that station. This information, plus other reactions of the sapsuckers, indicated the tentative locations of territories and nests. Each territory is occupied by only one pair of adult sapsuckers. The adults and their young occupy this territory until fall migration begins.

Later, I entered each territory and searched for the nest. The normal nesting activities of sapsuckers, and their willing response to imitation drumming, permitted efficient location of their nests.

### Observations

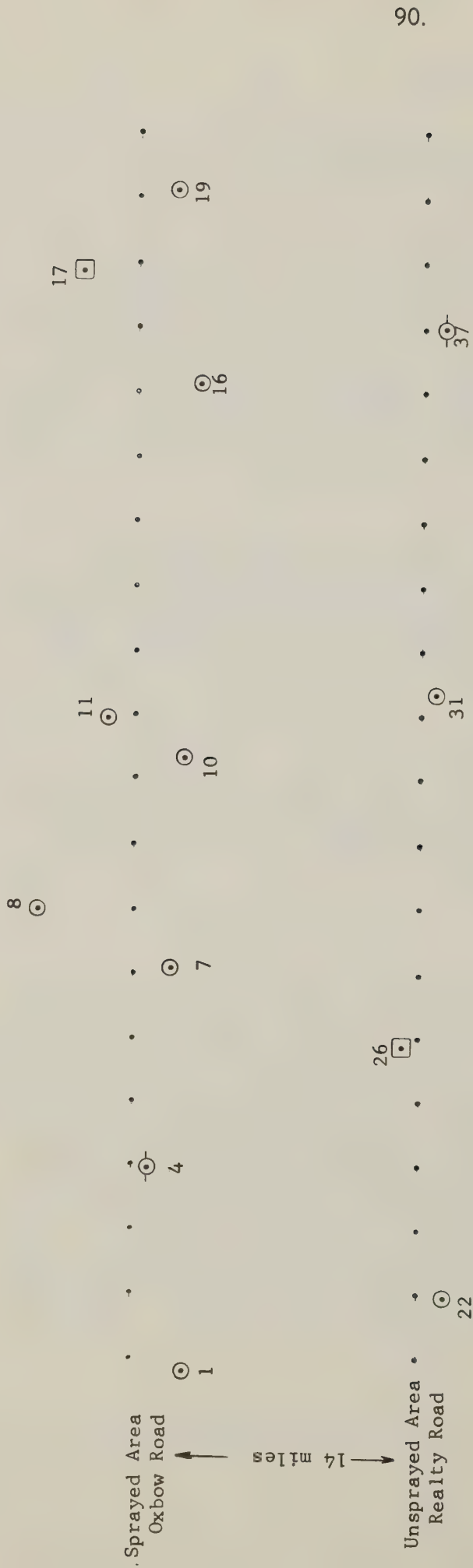
Observations of sapsucker activities were made before and after spraying, to help determine whether mortality or abnormal behavior occurred after the spraying. Two checks were scheduled for each nest within 48 hours after each of the two sprays. Other checks were made as time and situations permitted. If abnormal sapsucker activities were observed, these were explored further for clues to help determine whether they were caused by natural conditions or by the chemical.

Sapsucker activities were judged to be normal if they were in agreement with activities commonly performed during the current stage of sapsucker nesting, for example, pre-nesting, nesting, or post-nesting.\*

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\* Based on 1177 hours of personal observations of sapsuckers and their activities.

Figure 1. Location and status of sapsucker nests.





### Nestblocks

The effect of the chemical on survival of nestling birds is an important aspect of the observations. The sounds of sapsucker nestlings can easily be heard while they are in the nest-tree cavity and will verify that some birds are alive. But it also is desirable to compare the numbers of nestlings on the sprayed and unsprayed areas, to estimate whether normal numbers were produced and survived on the sprayed area. This was done by using the Nestblock Technique. The nest trees were cut when most nestlings were about 2/3 grown. A block about 36 inches long and containing the nest was cut from the tree. The nest was opened by a cut near the top of the nest cavity. Nestlings were banded and returned to the nest. The two pieces of the nestblock were fastened together. The nestblock was attached to a tree near the original nest tree and about six feet above ground. The calls of the nestlings for food soon led to resumption of normal feeding and care by the adults.

Nestblocks were prepared and nestlings banded between June 29 and July 6. Maine Forestry Department personnel assisted with all nestblock and banding work.

### Banding

Adults were not banded. Banding would have been desirable but there was insufficient time. However, when frequent observations are made at nests, loss of even a single unbanded adult is easily detected by distinct changes in nesting activities.

Nestlings were banded with a U.S. Fish and Wildlife Service metal band on one leg, and a plastic color band on the other leg. A different color combination was used for each territory.

### Cholinesterase

One complete brood of four nestlings were collected June 29 from nest 37 on the unsprayed area, and one brood of five from nest 4 on the sprayed area (Fig. 1). Dead birds were immediately frozen, and later were turned over to the U.S. Fish and Wildlife Service for brain cholinesterase determinations and comparison.

### Mortality

Mortality of an individual adult sapsucker was judged to

have occurred, if after the pair of birds were well-advanced with nesting: (1) One adult disappeared and was not present for a prolonged period, to participate in the current phase of nesting. (2) The surviving adult at first remained at the nest site and tried to maintain the nesting, but finally abandoned the current phase of nesting. (3) The surviving adult reverted to pre-nesting activities and tried to attract a new mate.

Mortality of nestlings was judged to have occurred if: (1) A dead nestling, or bones of a decomposed nestling were found in the nestblock. (2) A band was found inside the nestblock, with a leg bone inside the band. (3) Strong evidence that a predator destroyed the nestlings, and only the adult sapsuckers could be found in the territory.

### Other Wildlife

During the pre- and post-spray periods, all grouse and woodcock seen were recorded. Many observations of evening grosbeaks were also recorded. Several other species of birds and mammals were recorded when seen in the sapsucker territories.

### Results and Discussion

Detailed information was obtained for 74 sapsuckers, 53 on the sprayed area and 21 on the unsprayed area (Table 1). All recorded observations were made by one man, within the period May 19 through September 4. Field work was conducted in 32 days. The spraying apparently did not kill sapsuckers or seriously upset their normal nesting activities.

Table 1. Sapsuckers found on the sprayed and unsprayed areas

	<u>Sprayed</u>	<u>Unsprayed</u>
<u>1. Nests:</u>		
a. Found in May	9	4
b. Abandoned or destroyed	1	1
(1) Probable reason	Loss of female	Loss of female
c. Successful nests	8	3
d. All nestlings collected for cholinesterase analysis	1	1
e. All banded nestlings put into nestblocks	7	2
f. Successful nestblocks	7	1
<u>2. Adults:</u>		
a. Found in May	18	8
b. Disappeared	1	2
(1) Probable reason, taken by	Hawk	Hawk
c. Moved in to replace missing birds	Female	none
<u>3. Nestlings:</u>		
a. Produced by July 6	35	13
b. Collected for cholinesterase analysis	5	4
c. Banded and returned to nestblock	30	9
d. Mortality in nestblock	2	5
(1) Probable reason	Chilling	Predator
e. Matured and left nestblocks	28	4
<u>4. Banded juveniles seen after fledging:</u>		
Number of observations	29	1
(1) In nesting territories	7	1
<u>5. Total number of sapsuckers:</u>	53	21



### Nests

Nine occupied sapsucker nests were found in the sprayed area and four in the unsprayed area, between May 20 and 30. Locating the nests later permitted quick, positive observations, before and after spraying. All 13 nests were found before the first spray and there was ample time to determine that normal nesting was occurring.

Apparently no nestlings were directly exposed to either of the two sprays. When the first spray was applied, during early morning of June 4, eggs were being incubated at six nests, eggs were being laid at two nests (1 and 11), and the status of nest 17 was questionable. During the second spray, early morning of June 10, incubation was occurring at eight nests. Nesting was not occurring at nest 17; the male was present but the female was missing.

The sapsucker population in the sprayed area was twice as large as the population in the unsprayed area (Fig. 1). The old sapsucker feeding holes found on trees in all territories of the sprayed area suggests that the dense 1970 population was not unusual for that area, and that it had been similar for at least several years prior to 1970.

### Observations

Each territory in the sprayed area was visited an average of 16 times, 4 before spraying and 12 after. Territories in the unsprayed area were visited an average of 12 times. Observations were made anytime during daylight, the earliest being 5:27 A.M. and the latest 7:06 P.M.

The two post-spray inspections at each nest were made within 31 and 34 hours, respectively, following the sprayings on June 4 and 10. In the sprayed area normal nesting activities continued at eight of the nine nests, and these activities were identical with those that occurred in the unsprayed area. Among these 16 adults in the sprayed area, nothing was seen to indicate that sapsucker behavior had been affected by the sprays. These 16 adults remained alive, completed all phases of their nesting, and produced young birds that appeared and acted normal after fledging.

At the ninth nest in the sprayed area, some unseasonal sapsucker activities did occur and involved pair 17, but these apparently resulted from loss of the female several



An adult sapsucker enters its nest to feed nestlings. After spraying, spruce budworms were eaten by adults and were fed to nestlings, and other normal nesting activities were continued.



A sapsucker nestblock placed near the original nest tree. Adults soon resumed normal care of banded nestlings inside the nestblocks.



days before June 10. Other unseasonal behavior occurred near nest 16 soon after each spray. It involved the presence and actions of two strange sapsuckers in territory 16: a female at 11:18 A.M. on June 5, and a male at 1:15 P.M. on June 10. I believe the male was from nest 17. That deliberate lack of respect for the integrity of an occupied sapsucker territory was unusual for those dates. Otherwise, these birds appeared normal and healthy. A very strong possibility exists that male 17's territorial defensiveness and intensive efforts to secure a new female influenced this unusual behavior, but that suggestion does not satisfy all potential questions.

We cannot overlook the possibility that the chemical might have had some minor, temporary affect on several sapsuckers. However, if male 17 was affected, he remained vigorous and acted normally after June 10. He secured a new mate on June 12, having a choice of two surplus females; and they later produced eggs and nestlings.

The unseasonal behavior was observed only on a narrow strip involving nests 16 and 17, in the sprayed area (Fig. 1). Future monitoring should include observations to detect any repetition of this behavior.

#### Nestblocks and Banding

The adults resumed normal feeding and care of the nestlings less than 24 hours after the nestblock work was completed.

Eleven of the 13 nests were successful and produced 48 nestlings, 35 in the sprayed area and 13 in the unsprayed area (Table 1). Nine nestlings were collected for cholinesterase evaluation. Of the 39 nestlings banded and returned to their nestblocks, 32 survived, fledged, and apparently lived normally in their home territories.

Apparently the spraying did not reduce the number of nestlings produced. The average number of nestlings per nest was about 4.3 for both the sprayed and the unsprayed areas. Only one nest (8) had three nestlings, all others had four or five. In the sprayed area, most eggs were in the nests during both sprays. Hatching began soon after June 10, so the very small birds would have been exposed if any residual chemical occurred on insects or in the tree sap being fed to them. For example, nestlings were being fed in nest 10 on June 11; the nest contained four nestlings on June 29; all had left the nestblock by July 15; and on August 13 three of these juveniles were positively identified and were alive and acting normally. The fourth nestling probably was at a different sap tree.



### Cholinesterase

The brain cholinesterase levels of the nine nestlings sampled were nearly identical on the two areas, averaging 1.89 on the sprayed area (1.77 to 2.19), and 1.86 on the unsprayed area (1.73 to 1.90). This indicates that no effects of the chemical were apparent 19 days after the last spraying.

### Mortality

Very little mortality occurred among adult sapsuckers. In the sprayed area, the female disappeared from nest 17, between June 5 and 10. The evidence suggests that she might have been taken by a hawk. The evidence does not suggest that she might have been killed by the spray. Her male remained alive, and so did all other adults observed in the sprayed area. In the unsprayed area a female disappeared from nest 26, between June 5 and 10, and probably was taken by a hawk. Her male also disappeared from territory 26 sometime after June 12. He may have been taken by a hawk, or he may have been the live surplus male I found later near station 30.

Predation of eggs or nestlings apparently occurred before two nest trees were cut. After the females disappeared from nests 17 and 26, predators apparently destroyed the unguarded eggs. The second nesting at 17 failed when nestlings disappeared shortly after hatching.

The sprays apparently did not create a serious shortage of budworms or other insects needed by nestlings. Insects are an important food of sapsuckers. They fed heavily on spruce budworms before and after the sprays. Adults were observed either hunting, collecting, eating, or feeding the budworms to nestlings at six nests on the sprayed area, and at three nests on the unsprayed area. After the sprays, adults gathered vigorous budworms from the foliage and apparently-sick budworms hanging from the branches.

Survival or mortality of young sapsuckers within nests was verified after all living birds had left a nestblock. Nestblocks were opened and the contents carefully examined for evidence of mortality. Seven nestlings died in the nestblocks. This mortality probably was an indirect result of our nest disturbance and not caused by the chemical. If the chemical had been involved, other nearby nestlings should have died. On the sprayed area the two smallest nestlings died in nestblock 11: these exceptionally young birds were the smallest found, were only about one-fourth grown, and may have chilled before the adults resumed brooding and

feeding on June 30; the other three nestlings were larger and they survived. On the unsprayed area all five nestlings were killed by a predator that dug through decayed wood in the top of nestblock 22. All four parents of these nestlings survived.

After nestlings left the nestblocks, their survival was further verified. At least one banded juvenile was found living normally in or near its home territory in August or September. All were found in territory 19. All living juveniles probably could have been found if additional color banding and searching had been done.

Further evidence of normal nesting was indicated by the contents of the nestblocks, after all young had left. The contents were similar in the sprayed and unsprayed areas. Approximately similar quantities of droppings, insect remains, and wood chips were found in the nests. Those are typical, and they were similar in quantity to the nestblock contents I found near Orono, Maine in 1968.

Very little was seen of the adults after nesting was completed. Some adults frequented the trees where I found banded juveniles feeding, but I did not have time to search for other trees they used.

### Other Wildlife

I found no other dead or sick wildlife. Many birds and red squirrels were seen before and after the spraying, and their activities appeared to be normal. The following notes summarize some of my recorded observations.

Evening grosbeaks (Hesperiphona vespertina vespertina) were present in very large numbers on the spray area when I arrived May 19, and flocks of more than 50 birds were common. They remained abundant until at least mid-July, long after the sprays. They fed heavily on budworms, before and after the sprays. On July 1, large numbers flew eastward in the Oxbow Road area. By July 6, many budworm moths were flying. From July 6 to 16 grosbeaks were still present, but they were in family groups instead of the large flocks seen earlier. Many juvenile grosbeaks were seen, thus indicating that nesting occurred on the sprayed area. By August 11 - 13 and on September 2 - 4, grosbeaks were scarce along the Oxbow Road, and I assumed they had moved to other foods. Those few observed generally were flying high - as they do when migrating. At no time did I see many grosbeaks in the unsprayed area.

A hen woodcock (Philoheia minor) and one chick were seen May 29 in territory 10, the spray area. Between August 11 and September 3, I flushed four woodcock within 600 feet of that place. On August 11 two woodcock were seen in territory 1. Presumably these were local birds that had been exposed to the sprays.

A hen ruffed grouse (Bonasa umbellus), with at least six chicks were flushed in territory 17, on August 11. These were the only grouse I saw in the sprayed area. None were seen in the unsprayed area.

Ruby-throated hummingbirds (Archilochus colubris) were seen in the sprayed area before and after spraying. They commonly live near sapsuckers and make many visits for tree sap at holes made by sapsuckers. While searching for banded sapsuckers in August and September, hummingbirds were frequently seen visiting sap trees, in seven sapsucker territories in the sprayed area. I suspect that these were local hummingbirds and not migrants, which would indicate that these delicate birds survived the spraying.

Future pesticides monitoring might utilize either or both sapsuckers and hummingbirds. Both birds have characteristics, ranges, and habits that should make them exceptionally desirable for monitoring pesticides of various kinds, and over large regions.

### Conclusions

1. No evidence suggested that any of the 74 sapsuckers observed were killed or seriously harmed by the two applications of Accothion.
2. The techniques used for monitoring accomplished the objectives, and sapsuckers were a reliable indicator species. There is a substantial basis for future technique refinements. One man can accomplish all phases of the current techniques except one: he needs another person to help with the nest block preparation and the banding.
3. Hummingbirds may also have potential for monitoring. If so, sapsuckers and hummingbirds could be found and observed simultaneously.



### Recommendations

1. Refine pesticides monitoring techniques, to improve understanding and confidence in the results. Conduct monitoring in Maine in 1971 if there is spraying, and if not, refine the techniques without the use of chemicals.
2. Begin monitoring early in spring and continue it until fall migration.
3. Observe adult and juvenile birds after nesting, to insure that they survived the spray. Some adults should survive and appear later in their same territories, at least the year following the spraying, and possible longer. Then it would be possible to determine whether they were capable of conducting normal nesting the year following exposure to a chemical.
4. Do more color banding, to obtain additional information, and to increase confidence that individual birds survived after nesting ends. Use a color numbering code to identify individual juveniles. Band the adults, to facilitate observations in the current and later years.
5. Make more analyses of brain cholinesterase levels in nesting and adult sapsuckers. If hummingbirds are used for monitoring, consider cholinesterase analysis. If chemicals other than Accothion are used, consider appropriate analyses.
6. Survey the 1970 sprayed and unsprayed areas in May 1971, and determine whether the sapsucker populations remained normal the year after the spray.

## Studies of Forage, Milk, and Soil Samples

Kenneth B. Johnson

Supervisor of Dairies

Maine Department of Agriculture

### Objective

To sample grass forage, milk, and soil from a dairy farm within the spray area and test the samples chemically for presence of Accothion.

### Method

Samples of grass, milk, and soil from the Laurel Junkins dairy farm in Oxbow (See map in Appendix), were obtained on June 15 after completion of spraying in the area. The farm was well within the spray area but was not directly sprayed. The grass forage was obtained by severing it from the roots so that no soil would be involved. Five one-cubic foot samples of grass were obtained and kept separate in plastic bags.

Soil samples were taken by cutting a small circular area about one inch deep, removing any vegetation, and then taking one-half pint samples of soil from each five such dug areas. Five samples were taken and kept separate in plastic bags.

Two milk samples were taken from batches containing a mixture of three milkings. Samples were taken aseptically with a stainless steel ladle dipped into the milk tanks and were put into two sterilized plastic containers, each sample being one-quarter pint. The cows had pastured (fed) in two of the areas from which the grass was sampled.

All of the samples were submitted directly to the Maine Agricultural Experiment Station Laboratory in Orono, on June 16. Prior arrangements had been made for testing for the presence of Accothion by Stanley Getchell, Associate Professor of Chemistry. Between receipt of samples and chemically testing for Accothion, samples were held in freeze-storage at -25°C.

### Results

Findings are tabulated below.

<u>Sample No.</u>	<u>Item</u>	<u>Results</u>
4196	Soil	No Accothion found
4197	Soil	No Accothion found
4198	Soil	No Accothion found
4199	Soil	No Accothion found
4200	Soil	No Accothion found
4201	Grass	1.1 ppm. Accothion
4202	Grass	0.8 ppm. Accothion
4203	Grass	0.5 ppm. Accothion
4204	Grass	0.5 ppm. Accothion
4205	Grass	0.3 ppm. Accothion
4206	Milk	No Accothion found
4207	Milk	No Accothion found

### Discussion

Results of this study are interesting to compare with a similar study in a pilot test of Sumithion in 1968. Sumithion and Accothion are basically the same, difference in names being related to different manufacturers. Similar tests of forage, milk and soil at that time detected no trace of Sumithion.

### Conclusion

No objections are seen to the use of Accothion at this time. If it were used in the future involving food producing farms we would wish to monitor such areas for possible contamination.



APPENDIX

To: Dr. John Dimond

Report of Analysis....Accothion in Spray Material

Block Number	Date Sampled	Lab. Number	Accothion (fenitrothion) (percent)
7 Plot V	5 JN 70	R-13606	9.4
10 Plot IV	4 JN	R-13607	9.2
11 Plot III	4 JN	R-13608	9.6
12 Plot II	4 JN	R-13609	10.1
13 Plot I	4 JN	R-13610	4.9
12 Plot II	10 JN	R-13617	11.1
13 Plot I	10 JN	R-13618	10.5
7 Plot V	10 JN	R-13619	8.1
10 Plot IV	14 JN	R-13620	9.5
11 Plot III	14 JN	R-13621	9.0

29 June 1970

A. S. Getchell  
J. A. Blease



OXBOW, MAINE

